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Review article

Antibacterial biohybrid nanofibers for wound dressings

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ABSTRACT

Globally, chronic wounds impose a notable burden to patients and healthcare systems. Such skin wounds are readily subjected to bacteria that provoke inflammation and hence challenge the healing process. Furthermore, bacteria induce infection impeding re-epithelialization and collagen synthesis. With an estimated global market of $20.4 billion by 2021, appropriate wound dressing materials e.g. those composed of biopolymers originating from nature, are capable of alleviating the infection incidence and of accelerating the healing process. Particularly, biopolymeric nanofibrous dressings are biocompatible and mostly biodegradable and biomimic the extracellular matrix structure. Such nanofibrous dressings provide a high surface area and the ability to deliver antibiotics and antibacterial agents locally into the wound milieu to control infection. In this regard, with the dangerous evolution of antibiotic resistant bacteria, antibiotic delivery systems are being gradually replaced with antibacterial biohybrid nanofibrous wound dressings. This emerging class of wound dressings comprises biopolymeric nanofibers containing antibacterial nanoparticles, nature-derived compounds and biofunctional agents. Here, the most recent (since 2015) developments of antibacterial biopolymeric nanofibrous wound dressings, particularly those made of biohybrids, are reviewed and their antibacterial efficiency is evaluated based on a comprehensive literature analysis. Lastly, the prospects and challenges are discussed to draw a roadmap for further progresses and to open up future research avenues in this area.

Statement of Significance

With a global market of $20.4 billion by 2021, skin wound dressings are a crucial segment of the wound care industry. As an advanced class of bioactive wound dressing materials, natural polymeric nanofibers loaded with antibacterial agents, e.g. antimicrobial nanoparticles/ions, nature-derived compounds and biofunctional agents, have shown a remarkable potential for replacement of their classic counterparts. Also, given the expanding concern regarding antibiotic resistant bacteria, such biohybrid nanofibrous wound dressings can outperform classical drug delivery systems. Here, an updated overview of the most recent (since 2015) developments of antibacterial biopolymeric nanofibrous wound dressings is presented. In this review, while discussing about the antibacterial efficiency of such systems, the prospects and challenges are highlighted to draw a roadmap for further progresses in this area.

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1. Introduction

Globally, acute and chronic wounds, e.g. burns and diabetic ulcers, respectively, impose a notable burden to patients and the healthcare systems. In Europe, there are more than 55 million patients suffering from diabetes, thereof 8 million ones are vulnerable to developing a diabetic foot ulcer. As a consequence of inefficient treatment of such ulcers, annually up to 450,000 lower limb amputations take place that can cost as much as €2–2.5 billion [1]. The statistic for the US indicates 6.5 million patients whose annual treatment cost is as much as $25 billion [2]. In the case of acute wounds, in the USA, trauma induced wounds that are the most incident ones result in nearly 41 million emergency department referrals and 2.3 million hospital stays. This situation imposes large relevant costs of $670 billion / year including those of health care as well as disability [3]. To alleviate such a costly burden, there is extensive research in progress to develop technologies able to treat wounds effectively in a short time. Appropriate treatment strategies include use of wound dressings with a high healing rate that can hinder infection, prevent the undesired outcomes and lead to reduction of costs.

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Skin wound dressings are a crucial segment of the wound care industry and trade worldwide. The global market of these products is estimated to surpass $20.4 billion by 2021 from $17.0 billion in 2016, due to the rising aging population and increasing incidence of chronic diseases such as diabetes [4]. The wound dressings currently available in the market are typically in the form of hydrogels, films, sponges, and foams. As a relatively new class of wound dressing materials, nanofibers have emerged that offer distinct advantages. Nanofibrous meshes comprising many intersecting nanofibers as small as a few microns to a few hundred nanometers can provide a notably large exposed surface area and nanoporosity, thereby facilitating interaction with the cells available in the wound bed through an extracellular matrix (ECM) mimicking structure [5].

While nanofibers can be produced via several methods such as drawing [6], template synthesis [7], phase separation [8] or self-assembly [9], electrospinning is a superior technique due to its simplicity, cost efficiency, and versatility [10–16]. The drawing method, fabricating individual long nanofibers, is restricted to viscoelastic materials able to tolerate large deformations while remaining cohesive to resist against pulling stresses. Employing a nanoporous template, the template synthesis allows for production of solid/hollow yet discontinious individual nanofibers. Moreover, the time consuming phase separation method comprises stages of dissolution, gelation, extraction by another solvent, freezing, and drying, leading to formation of a nanoporous foam. Self-assembly methods are also time consuming techniques wherein single, building blocks are arranged as particular configurations to offer desired functions [17].

In comparison to the golden benchmark for wound dressings including films, foams, and microfibrous and mesh ones, nanofibrous dressings usually exhibit higher porosity thus allowing more efficient permeability for water and oxygen, and superior interchange of nutrients as well as exclusion of metabolic wastes [18]. Also, nanoscale fibers, conferring the dressing small interstices and high surface area, can enhance hemostasis. Not only the small pore size of nanofibrous dressings protects the wound against bacteria infection and cell/tissue ingrowth, also advantageous over the microfibrous and mesh commercial counterparts, nanofibrous dressings can provide excellent conformability, thus a better coverage and protection of the wounds from infection [19]. Most significantly, compared to commercial dressings, the expansive surface area of nanofibrous dressings enables efficient loading/incorporation of drugs. Thus, they show a promising potential for development of advanced, biologically active dressings. Such capability of nanofibers has become highly attractive in the biomaterials sector leading to a vast research effort, as documented by several relevant reviews in recent years [20–31]. Different to those publications, specifically, we focus here only on biohybrid nanofibrous wound dressings based on biopolymers combined with antibacterial agents in diverse modes. These advanced classes of antibacterial nanofibrous dressings can potentially replace their established counterparts, that are antibiotic delivery nanofibrous systems, given the expanding concern about the increase of antibiotic resistant bacteria. In this context, depending on the type of the antibacterial agent incorporated, being nanoparticles, organic macromolecules (peptides or amino acids), and nature (e.g. plant) derived compounds, nanocomposites, biofunctionalized, and blend systems, respectively, are introduced. As a specific highlight, in this review we evaluate the most relevant studies published since 2015 and unravel the latest status of research in this area.

2. Wound healing

The proper design of a skin wound treatment strategy, including the application of dressing systems, is intimately associated with having vast, detailed knowledge about the wound healing process and the involved factors and local conditions affecting the wound.

As soon as a superficial wound forms, several mechanisms start to operate to clean up the wound milieu from the invading materials and to repair the skin primary structure. This complex healing process engages diverse inflammatory cells, cytokines, chemokines, nutrients, and matrix molecules, that are released into the wound bed. In general, wound healing comprises three principal steps namely; inflammation, proliferation, and remodeling [32]. Fig. 1 shows the main mechanisms of wound healing and the elements taking part in the process. The inflammatory phase commences right after completion of hemostasis and thereby pathogens and foreign materials are removed from the wound milieu. To do this, vascular permeability is provoked by vasodilation, enabling concentration of neutrophils and monocytes within the wound bed [32]. This step is also modulated through an intricate interplay of cytokines, leading to transformation of monocytes to macrophages [33]. The as- differentiated macrophages play two important roles: 1) phagocytosis and digestion of the tissue debris and neutrophils, and 2) release of cytokines and growth factors stimulating cellular proliferation and migration. The proliferation step starts after almost 3 days and based on activities of fibroblasts and generation of ground substance and collagen. The fibroblasts present in the area and those derived from blood proliferate and migrate, thereby creating the wound granulation tissue and also a new ECM. Additionally, a number of fibroblasts are converted (differentiated) to myofibroblasts to facilitate wound closure [34]. The as-formed ECM has a decisive effect in the modulation of cells performance during the wound healing process [35] through molecular signaling (mainly by integrins). Later, cellular activities including proliferation, differentiation, or even apoptosis are triggered by such signals [36]. Keratinocytes and fibroblasts are main skin cells that release integrins, being influenced by the ECM modulation activity [37]. Also, the ECM proteins regulate the behavior of cytokines and growth factors including transforming growth factor-β (TGF-β) and platelet-derived growth factor (PDGF) released by macrophages and activated platelets, respectively [38,39]. The mentioned regulative role of ECM in cellular activities is further supplemented by its fibrous protein (made of collagens, elastin, and fibronectin) supportive structure. Cells anchor on such a fibrous architecture, a vascular network is formed therein, and the growth factors are protected against degradation [39]. During the course of the proliferation phase, endothelial cells quickly grow and induce angiogenesis inside the granulation tissue. The transition of wound state to a remodeling (maturation) state takes place after about 2–3 weeks [40]. The maturation of the wound tissue leads to further cross-linking and thereby formation of normal tissue. Upon accomplishment of maturation, the vascular network immediately returns to its former less developed state [41]. As shown in Fig. 1, wound healing can be accelerated by innervation [42].

When an acute wound, e.g. by surgery, cut, trauma, etc., forms, the above mentioned processes of hemostasis, inflammation, cell proliferation, and tissue remodeling are triggered [43]. Any abnormality in the healing stages can lead to chronicity or fibrosis. Such deviation could originate from an existing disease affecting e.g. blood circulation (peripheral vascular disease) and the immune system (immunosuppression). It could also be driven by a metabolic disorder (e.g. diabetes), consumption of particular medications, or radiation therapy [32]. In contrast to acute wounds, chronic ones are unable to follow the physiological steps to heal in a short period of time [34]. Chronic wounds show more notable protease activity, lower growth factor activity, and larger amount of pro-inflammatory cytokines, than acute wounds do [44]. As shown in Fig. 1, chronic wounds can be readily subjected to infection and thereby show a persistent abnormal inflammation

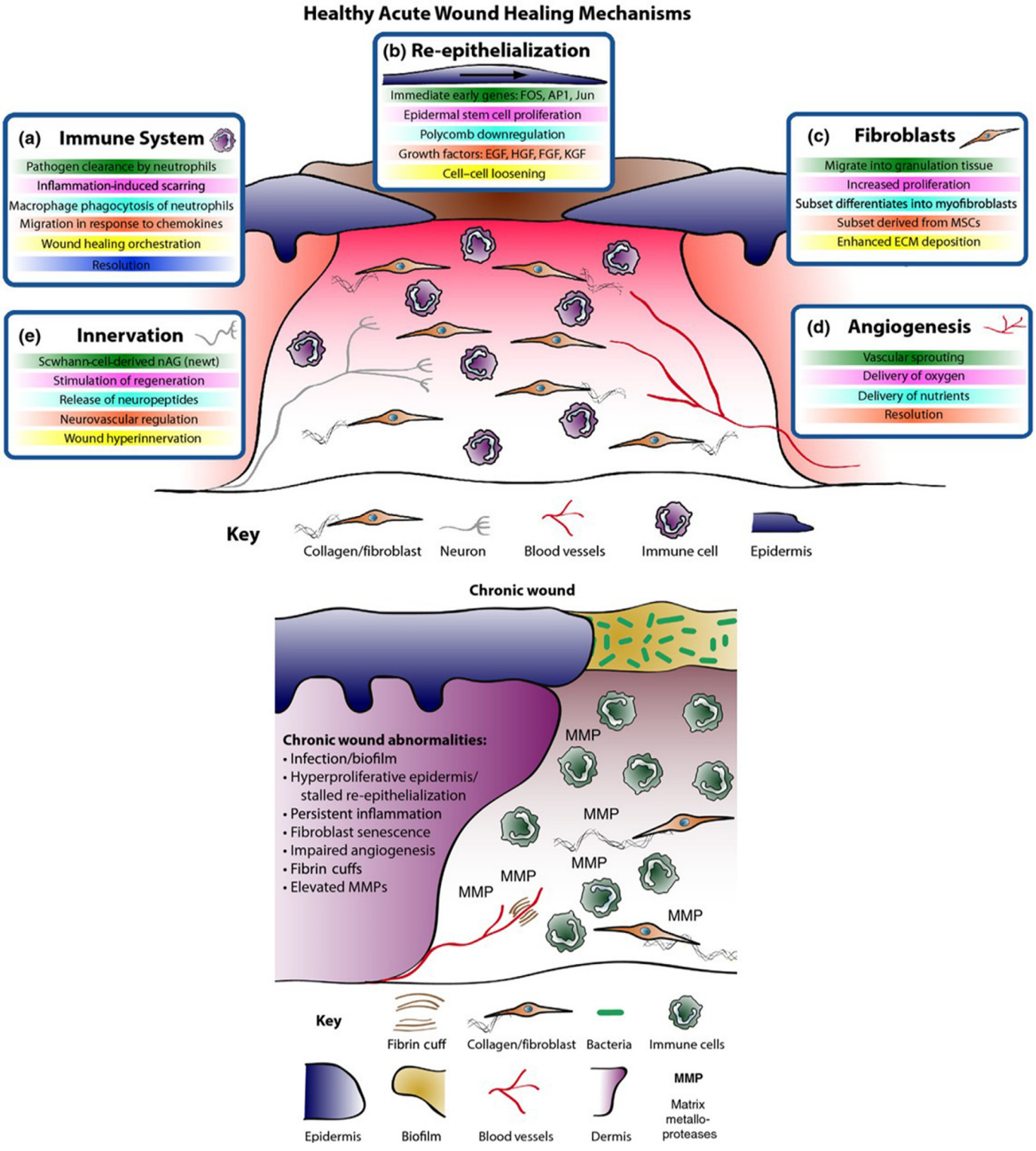

Fig. 1. The healing process for acute wounds (Top) and the chronic wound biology (Bottom). EGF, epidermal growth factor; HGF, hepatocyte growth factor; FGF, fibroblast growth factor; KGF, keratinocyte growth factor; MSC, mesenchymal stem cell; nAG, newt anterior gradient protein. Reproduced with permission [34]. Under a CC-BY License, Copyright 2015, John Wiley and Sons.

response. Re-epithelialization is inhibited while keratinocytes existing in the wound stay hyperproliferative. Moreover, an incomplete granulation tissue forms that does not support the healing process. Such a deficiency stems from the increased density of matrix metalloproteases (MMPs) as well as insufficient infiltration of fibroblasts. Also, neovascularization does not happen adequately and fibrin cuffs hinder the available vessels to supply oxygen throughout the wound, thereby causing the wound to be hypoxic [34].

3. Treatment of chronic wounds

There are two major therapeutical choices for chronic wounds that, however, never guarantee complete recovery. While the nega-

tive pressure wound therapy is able to cure only very small chronic wounds, the hyperbaric oxygen therapy could fail even after a therapeutic period of one year [45]. In addition to such therapies, to encourage wound healing, the use of growth factors has been appealing and many researches have been conducted on them during the past few decades. Yet, among the many studied growth factors, only a few (e.g. PDGF) have shown a moderate healing efficiency in a double-blinded arbitarilly controlled study [46]. Infection is regarded as the most well-known barrier against wound healing. To address such a challenge for local wound infections, topical antimicrobials and cleansing agents have been applied since long time ago. Despite their popularity, it has been reported that regular application of antibiotic ointments such as those containing gentamicin and neomycin not only does not improve the wound status but also causes discomfort to the patient, alongside contact dermatitis and antibiotic resistance [32]. Low concentrated formulations of povidone iodine have shown optimum antimicrobial efficiency for a large range of bacteria, while being nondetrimental to cellular activities. Manuka honey in medical grade is another antimicrobial agent with peroxide and nonperoxide antibacterial performance. This agent is able to inactivate over 50 kinds of bacteria [47] and can be applied either as a topical preparation or as honey-impregnated dressings [48].

Skin substitutes comprising a bioderived substance coupled with a material allowing for its secure positioning within the wound, are another option for management of chronic wounds. However, such products can be expensive and this hinders their extensive application. There are currently several commercial skin substitutes based on porcine collagen or polypeptide coated mesh materials or a porcine xenograft [32]. Another relevant option is a skin substitute composed of newborn foreskin tissue derived fibroblasts, a bioresorbable polyglactin mesh, and extracellular matrix. In this material, the wound healing is accelerated by the fibroblast performance in production of cytokines, growth factors, collagen, and glycosaminoglycans. Such a system has proved efficient for treating burn wounds and venous/pressure ulcers [49]. However, the cost efficiency of its application is not certain yet. One very important means in managemnet and control of chronic wounds is the development of effective wound dressings.

Wound dressings are made to protect the wound, remove exudate, inhibit exogenous microorganism invasion and improve appearance [50]. The most promising wound dressings are biocompatible, enable physical protection of the wound milieu against penetration of bacteria, are highly porous, thus air permeable, and drive epithelization [51].

The first, ancient examples of wound dressing materials have been found in the Sumerians civilization where they used poultices of mud, milk, and plants to enable wounds to heal. The Egyptians also created bandages made of honey plasters, plant fibers, and animal fats to cover and treat wounds. In the chronological trend of development of wound dressings, the revolutionary breakthrough came up within the course of the 19th century with the progress in microbiology and cellular pathology [52]. In this regard, in the 1960s it was uncovered that moisturizing a wound notably helps it heal. This fact was based in the development of the new generation of wound dressings [52]. In addition to a moist atmosphere, an efficient wound dressing needs to remove excessive exudates, to supply thermal insulation, to shield the wound bed against mechanical trauma and bacterial invasion, to permeate gases and fluids, to be peeled off with no damage to the wound, and to be safe in terms of cytotoxicity and allergic reactions [53].

There are five main classes of moisture retentive wound dressings including: films, foams, hydrocolloids, alginates, and hydrogels (Table 1) [48]. The polymeric semi-permeable films (e.g. Tegaderm) are elastic, thin, and transparent polyurethane (PU) sheets that stick to skin using acrylic. Foams in fact comprise a hydrophobic PU foam layer whose surface is coated by a hydrophilic material to hinder drainage leakage and to hamper bacterial invasion [48]. Foams (e.g. 3 M adhesive foam) also show an absorptive feature and can be applied for exudative wounds [32]. Hydrocolloid dressings are soft and conformable and consist of an adhesive carboxymethylcellulose, gelatin, and pectin matrix with a foam or PU film support layer. In addition to their moisture retention ability, they interact with wound exudate and create a yellow gel, facilitating autolytic debridement [48]. Hydrocolloids block air passage and have a long lifespan. Yet, due to their impermeability, they are inefficient for exudative wounds [32]. The alginate nonwoven dressings (e.g. Algisite), whose fiber composition is derived from seaweed, absorb extensive amounts of fluids, thus are typically used for notably exudative wounds [32]. Alginate dressings are able to swap calcium for sodium, thereby further absorbing exudate and also imparting hemostasis [48]. In relation to commercial fibrous wound dressings, Mo-Sci corporation (Rolla, MO, USA) has produced a fibrin cloth mimicking fibrous dressing made of biodegradable borate glass fibers. The fibers that are rich of calcium provoke epidermal cells migration and thus wound healing [54]. On the other hand, hydrogels are cross-linked polymeric networks that can contain up to 96% water. They are applied either as sheets or liquid gels that cover a wound or are injected into a wound, respectively. The specific features of hydrogels are promising for dry, necrotic wounds. Particularly, they can offer a soothing and cooling effect and thus relieve pain [55].

Wound dressings are promising options for control and management of chronic wounds. With the evolution of nanotechnology, nanostructured dressing materials are gaining increasing interest and attention. Particularly, nanofibrous wound dressings exhibiting biomimicry and other structural merits previously mentioned are of paramount importance. The beneficial topographical features can be supplemented with biological cues to improve cellular recognition when biopolymers are used as nanofiber materials.

4. Biopolymeric nanofibrous wound dressings

Taking into account the promising biocompatibility and ecofriendliness of biopolymers, many biopolymeric nanofibers have been studied for construction of efficient wound dressings. Originated from green, renewable resources, biopolymers, are popular for regenerative medicine. This popularity stems from their inherent bioactivity and biodegradability and resemblance to the ECM in terms of structure and surface topography [56]. Among the biopolymers used for wound treatment, polysaccharides are the most studied ones. These carbohydrate polymers comprise long molecules of monosaccharide units, successively arranged through the glycosidic bonds. These include cellulose, hyaluronic acid, chitin, chitosan, and alginates. This list of biopolymers is further completed by addition of collagen, silk fibroin, dextran, elastin, that have shown applicability as biocompatible wound dressing materials [5,43,57–59]. Table 2 summarizes the main types of biopolymers studied for wound healing purpose alongside their pros and cons.

Cellulose is a largely available polysaccharide made of glucose that has drawn attention for wound healing. As a wound dressing for chronic wounds, it alleviates pain and accelerates the healing process. In relation to the partial- and full-thickness wounds, cellulose dressings drive the granulation and epithelialization processes, thus assisting tissue regeneration. As an example, a wound dressing composed of wood based nanofibrillar cellulose has been produced to treat burn patients [60]. This cellulose based dressing induces a faster epithelialization than its commercial counterpart. Other than cellulose derived from plants, cellulose is also synthesized by bacteria (*Acetobacter xylinum* (Acetobacteraceae)), called

Table 1
Main classes of the moisture retentive wound dressings (Reproduced with permission [48], Copyright 2016, Elsevier).

Wound dressing type	Description	Pros	Cons	Commercial names
Films	Elastic, thin PU layers with an acrylic adhesive; suitable for donor sites for split-thickness skin grafts	Protect the wounds against invasion of bacteria, are gas permeable, and due to transparency enable visual check of the wound	Insufficient removal of fluid, and the fluid drainage could be even destructive for the newly formed epithelium	Tegaderm (3 M Healthcare), Polyskin II (Kendall Healthcare), Bioclusive (Johnson & Johnson Medical), Blisterfilm (The Kendall Co), Omniderm (Omikron Scientific Ltd), Proclude (ConvaTec), Mefilm (Mölnlycke Health Care), Carrafilm (Carrington Lab), and Transeal (DeRoyal)
Foams	Bilayer dressings comprising a hydrophobic PU foam surface coated with a hydrophilic material; suitable for the wounds located over bony areas, inside body cavities, and the wounds with low and moderate exudate production	Moisture absorption and retention, blocks drainage leakage and bacterial invasion, and readily conforms with the wound area	Possibly sticks firmly to the wound area in case of drainage desiccation	Polymem (Ferris Corp), Allevyn (Smith & Nephew United), Biopatch (Johnson & Johnson Medical), Curafoam (The Kendall Co), Flexzan (Dow Hickam), Hydrasorb (Tyco/Kendall Co), Lyofoam (ConvaTec), and Mepilex (Mölnlycke Health Care)
Hydrocolloids	Pliable sheets consisting of water-resistant foams or gels within PU films; suitable for the wounds with poor exudate production	Encourage formation of granulation tissue, are easy to use, and water-resistant	Undergo a gelation process, drainage, and not recommended for cavities	Duoderm (ConvaTec), NuDerm (Johnson & Johnson Medical), Comfeel (Coloplast Sween, Inc), Hydrocol (Dow Hickam), Cutinova (Smith & Nephew), Replicare (Smith & Nephew United), and Tegasorb (3 M)
Alginates	Composed of algae and kelp derived polysaccharides; suitable for exudative wounds	Absorptive, induce hemostasis, and applicable for sinuses	Not usable for dry wounds; necessitate regular dressing changes for the highly drainage producing wounds	Algiderm (Bard), Algisite (Smith & Nephew), Algisorb (Calgon-Vestal), Algosteril (Johnson & Johnson Medical), Kaltostat (ConvaTec), Curasorb (The Kendall Co), Sorbsan (Dow Hickam), Melgisorb (Mölnlycke Health Care), SeaSorb (Coloplast), and Kalginate (DeRoyal)
Hydrogels	Hydrophilic Cross-linked polymer networks with high water retention ability; suitable for dry, necrotic wounds	Enable autolytic debridement and confers a comforting effect for the patient	Possible skin maceration regarding highly exudative wounds	Vigilon (CR Bard), Nu-gel (Johnson & Johnson Medical), Tegagel (3 M), FlexiGel (Smith & Nephew), Curagel (The Kendall Co), Clearsite (Conmed Corp), Curafil (The Kendall Co), Curasol (The Kendall Co), Carrasyn (Carrington Laboratories), Elasto-Gel (SW Technologies), Hypergel (Scott Health Care), Normgel (SCA Hygiene Products), 2nd Skin (Spenco Medical, Ltd), and Transigel (Smith & Nephew)

PU: polyurethane.

Table 2
Diverse biopolymers suggested for construction of wound dressings.

Biopolymer	Pros	Cons	Reference
Collagen	Induce native biomolecular cues correlated with cellular activities including adhesion, proliferation, and migration as well as immune reactions	Processing stimulated denaturation, poor mechanical properties and high degradation rate, and possible prior and viral contamination in the case of the collagen extracted from animal tissues [56]	[5,70,71]
Gelatin	Biocompatible and biodegradable, with low antigenicity [5]	Intricate processing and harvesting [72]	[73]
Chitosan	Biocompatible, negligibly toxic, structurally resembling natural glycosaminoglycans, and degradable by enzymes (chitosanase and lysozyme) [5,56]	The purity, origin, and molecular weight distribution are crucial in process characteristics and the nanofibrous mesh architecture. Likely danger of disease transmission and antigenicity [72]	[74]
Fibronectin and fibrin	Provokes cell adhesion and spreading, fibrin hydrogel shows high tissue-like water capacity, adjustable mechanical properties matching those of soft tissues [56]	fast *in vivo* degradation rate and poor structural integrity during application [56]	[75]
Alginate	biocompatibile, negligibly toxic, nonimmunogenic, inexpensive, and simply cross-linked [56]		[76]

bacterial cellulose. This type of nanostructured cellulose shows promising physicochemical and mechanical characteristics, is biocompatible and biodegradable, and has a notable hydration and bactericidal potential [59]. Mimicking the ECM structure, bacterial cellulose can be widely employed for tissue engineering and in reconstruction of damaged tissues, for wound healing as well for blood vessel regeneration [61]. As a more sophisticated version, cellulose dressings can be upgraded by either blending with other biopolymers or incorporation of a wide range of active additives including enzymes, antioxidants, hormones, vitamins, and antimicrobial drugs [57]. For instance, cellulose acetate nanofibers have been functionalized by the incorporation of gallic acid, a polyphenol compound with anti-inflammatory, antibacterial, and antioxidant activity and commonly found in vegetables, nuts, tea leaves, and fruits [62]. Such biohybrid nanofibers show an optimum bactericidal effect for *Staphylococcus aureus*, thus high potential for wound dressing applications.

Hyaluronic acid (HA) is a glycosaminoglycan commonly present in vertebrates' conjunctive tissues, e.g. bone and skin, and is one of the most important constituents of the ECM. In terms of composi-

tion and structure, HA is a natural nonimmunogenic linear polysaccharide composed of N-acetyl-D-glucosamine and glucuronic acid. It is promptly biodegraded when used within the human body [63]. There are several classes of HA based hydrogels proposed for wound healing, based on glycidyl methacrylate [64], thiol [65], and DNA [66] functionalized HA whose functionality promotes their networking. As a nanofibrous wound dressing material, HA largely absorbs exudates and supports the cellular migration and proliferation thus facilitating regeneration of the lost tissue and wound healing [67]. Shin et al. [68]. developed a fibrous wound dressing consisting of HA core- poly(lactic-*co*-glycolic acid, PLGA) shell fibers loaded with epigallocatechin-3-*O*-gallate (EGCG). As challenged in streptozotocin-induced diabetic rats, the HA/PLGA-E fibrous wound dressing accelerated the healing process of diabetic wounds and regenerated skin. HA/poly(vinyl alcohol) (PVA) blend nanofibers have also been produced for wound dressing [69]. In such a system, PVA acts as the carrier polymer for HA and additionally hydroxypropyl-βcyclodextrin (HPβCD) is coupled as a stabilizer for electrospinning, allowing for a water based nanofabrication process.

Chitin and chitosan (CS) show remarkable biodegradability and biocompatibility and are considered outstanding options for development of wound dressings. Chitin is in fact the largest available natural amino polysaccharide (poly-N-acetyl-glucosamine) with an annual production volume equal to that of cellulose and typically traced in the exoskeleton of the invertebrates, insects, and crustaceans as well as in the fungi cell wall [57]. Chitosan is a poly-N-acetyl-glucosamine and consists of arbitrarily arranged β-(1→4)-D-glucosamine and *N*-acetyl-D-glucosamine. It is derived from chitin, through its alkaline deacetylation e.g. by exposure of shrimp shells to NaOH. Proportional to the deacetylation extent, the properties of chitosan differ and therefore in relation to wound healing it can be implemented as a proliferation promoter, antibacterial agent and macrophage activator [77]. A diverse range of formulations based on chitosan has been suggested that mostly involves hybrids and composites comprising other supplementary compounds such as sodium alginate, fibrin, pectin, epidermal growth factor (EGF), gelatin, ciprofloxacin, cellulose, silver and ZnO nanoparticles, bioactive glasses, tricalcium phosphate, thyme oil, Aloe vera, among others [42,57,74,78–80]. These compositions are realized as nanofiber, film, microbead, sponge, hydrogel, and mesh membrane dressing systems. As antibacterial CS based nanofibrous wound dressings, several formulations such as honey/polyvinyl alcohol (PVA)/CS [81], bacterial cellulose/CS/polyethylene oxide (PEO) [82], PVA/CS/tetracycline hydrochloride (TCH) [42], have been proposed. Regarding the CS nanofibers and their biomedical applications, interested reader is referred to a very recent review paper [83].

Alginate is a nature-derived anionic polymer that is extracted from brown seaweed and shows optimum biocompatibility and negligible toxicity and is inexpensive [84]. From chemistry standpoint, alginates are characterized as linear branchless polysaccharides comprising various contents of (1→4′)-linked β-D-mannuronic acid and α-L-guluronic acid subunits. By tailored processing of algae, extremely absorbing natural textures made of calcium alginate, calcium–sodium alginate, collagen–alginate, and gelatin–alginate are achieved that can potentially be applied as wound dressings [57]. For wound healing application, it preserves a proper level of moisture within the adjacent microenvironment, notably reduces bacterial infection at the wound bed, and drives wound healing [85]. Given the high porosity and lack of adhesiveness of the alginate dressings, an additional dressing seems to be necessary for the sake of protection and stabilization [86]. As a nanofibrous wound dressing, alginate has been blended with synthetic polymers including PVA [87], facilitating electrospinning, and antibacterial agents such as honey.

Proteins such as collagen, which is the largest available protein in the human body, are also commonly used to treat wounds [88]. Produced by fibroblasts, collagen promotes the cellular and molecular cascade taking place within the course of wound healing, tissue regeneration and wound debridement [89]. Additionally, providing a sacrificial material, dressings made of collagen are able to overcome the concern of high concentration of matrix metalloproteinases (MMPs), typically released in chronic wounds, that deteriorate nonviable and viable collagen of the human body [90]. Derived from animal origins of bovine, porcine, and avian, collagen dressings are beneficial for recovery of partial and full-thickness wounds with a low up to average level of exudates [57]. In this regard, Zhou et al. [58] synthesized tilapia skin extracted collagen nanofibers that could stimulate human keratinocyte (HaCaT) cell activities and epidermal differentiation, thereby accelerating the wound healing rate. As a conventional wound dressing, collagen fibrils are coupled with other natural and synthetic polymers to enable wound exudate absorption and preservation of moisture to boost wound healing [3,91]. For instance, salmon milt DNA, anionic polysaccharides, minocycline based hydrogels, α-tocopherulate, and alginic acid are some of the compounds combined with collagen for the purpose of construction of wound dressing material [57]. With an almost same composition and biological characteristics of collagen, Gelatin (GE) made through denaturation of collagen offers less antigenicity. Having the arginine–glycine–aspartic acid (RGD) sequences of collagen, gelatin optimally enables cell adhesion. Also, it includes high extents of glycine, proline, and hydroxyproline that promote and expedite healing of damaged soft tissues and wounds [92,93]. Gelatin is also commonly hybridized with other natural additives such as alginate, EGF, and hyaluronate to realize optimum wound dressing formulations [57]. A gelatin/keratin blend nanofibrous dressing was developed by Yao et al. [93] that could optimize cell proliferation, adhesion, and migration, thus promoting wound healing and also vascularization, as witnessed through an animal model test.

Derived from mulberry silkworm *Bombyx mori*, silk fibroin (SF) is a natural biopolymer that has been widely employed for diverse biomedical applications, thanks to its favorable biocompatibility, biodegradability, negligible inflammatory response, inexpensiveness and green processing [94]. In addition to such promising characteristics, due to adherence, pliability, and high exudate absorption capacity, SF have found application as a wound dressing material. This natural polymer is processed either alone or as a hybrid involving, e.g. chitosan, multiwalled carbon nanotubes, alginate, etc. [57]. SF could be achieved from nonmulberry silk derivatives such as *Antheraea assama* and *Philosamia ricini*, as well [43]. The protein sequence of this version of SF contains RGD motifs, that can ease attachment to the cells' integrin receptors [95]. As a result, such a feature enables optimal cell-material interactions and drives the wound repair process. SF is optimally able to mimic the skin milieu, minimize scarring and reduce atopic dermatitis, thus supporting wound healing [96]. Such beneficial characteristics have encouraged researchers to develop wound dressing systems based on SF and its derivatives [97,98]. Selvaraj and Fathima [99] fabricated antioxidant Fenugreek/SF nanofibrous wound dressing that could induce wound healing alongside full re-epithelialization and collagen deposition. Detailed information regarding SF based wound dressing are available in previously published comprehensive reviews [100,101].

Though the biopolymeric nanofibers *per se* offer interesting potential for wound healing, they alone are rarely able to satisfy both concerns of wound healing and disinfection adequately. Thus, they must be hybridized with functional agents that promote healing rate and are bactericidal. Specifically, to address the challenge of infection in the chronic wounds, a wide range of drugs, antibacte-

rial agents, e.g. Ag nanoparticles and herbal derivatives, e.g. honey and essential oils, have been hybridized with the nanofibers.

5. Antibacterial biohybrid nanofibrous wound dressings

The wound dressings with antibacterial activity prevent wound infection that is indeed a barrier against normal wound healing process. Typically, chronic wounds are readily subjected to bacteria and microorganisms that notably provoke inflammation and hence challenge the healing process [5]. Infection under the dressing extends the inflammation response, impedes re-epithelialization and collagen synthesis, and delays the healing process, thereby increasing hospital stay and treatment costs [102]. Infection is caused by the microorganisms, transferred from the hospitalized patients, internal resources and the surrounding skin [103]. Adhesion of bacteria onto a wound surface leads to formation of biofilms, that are the rich bacteria containing zones, potentially shielding them against the immune system and antibiotics. As a result, endotoxins are liberated that engender sepsis, and eventually death [104]. Thus, wounds need to be treated with wound dressings containing antibiotics and antibacterial agents to avoid bacterial infection and biofilm formation in the wound milieu. In this context, antibiotics e.g. penicillin and methicillin have been included in both skin wounds and dressing materials [97]. Yet, evolution of antibiotic enduring bacteria has somehow impacted the use of traditional antibiotics. For this reason, other sorts of antibacterial agents such as quaternary ammonium compounds, metallic ions, nanoparticles, and antimicrobial polymers can be suggested. These classes of antibacterial materials typically suffer from either insufficient efficiency or induction of cytotoxicity effect [97,105]. In the last couple of years, a new generation of antimicrobial agents called antimicrobial peptides (AMPs) have emerged. Such bio-derived antibiotics are found in diverse creatures such as mammals, fishes, insects, amphibians, and surprisingly in some bacteria, and help the host remain immune against bacteria, fungi, or viruses [106]. Despite having different structures, AMPs mainly possess cationic and amphiphilic zones with α-helical conformation, that can potentially damage the bacterial cell membrane [107]. AMPs are able to kill a large range of bacteria quite promptly with a remarkable efficacy [108]. Moreover, AMPs have proven efficient in wound healing through enhancing re-epithelialization and angiogenesis, neutralizing LPS, and immunomodulation [109,110].

While some biopolymers such as chitosan have shown inherent bactericidal effect [111], other types of polymeric wound dressings need to be equipped with antibacterial agents to minimize the adverse effects of infection. Fig. 2a and b schematically show a polymeric wound dressing with antibacterial activity, that not only physically shields the wound against bacterial attack but also facilitates the migration and differentiation of fibroblasts.

In the case of nanofibrous wound dressings, depending on the type of the antibacterial agent and the mode of loading on the nanofibers, several categories of antibacterial nanofibrous wound dressings have been envisaged and studied. Other than the antibacterial systems delivering drugs (drug delivery), in case the antibacterial agent is incorporated into or blended with the main biopolymer or surface resided on the biopolymer nanofibers using a bioconnector, different classes of nanocomposite, blend and biofunctionalized antibacterial nanofibrous wound dressings, respectively, are defined.

5.1. Antibiotic delivering antibacterial biohybrid nanofibers

As a new generation of interactive dressings able to cure bacterial infection, research has been directed towards synthesis of antibiotic loaded nanofibrous dressings. Such structures enable localized treatment of wounds with antibiotics and antimicrobials. Particularly, for chronic wounds including diabetic foot ulcers, due to poor blood circulation at the extremities, antibiotics should be delivered locally for a short time rather than via systemic administration. Thus, a topically administered drug therapy is more effective and selective to the wound and side effects are minimized [112].

With an extremely large surface area, nanofibers are promising candidates for acting as a drug carrier. Many antibiotics and anticancer drugs have been conveniently incorporated into electrospun polymeric nanofibers for local delivery [113]. Traditionally, incorporation of antibiotics into nanofibers is done via blending them into the polymer, followed by electrospinning of the blend or by core-shell electrospinning wherein the antibiotic is located within a polymeric outer shell, Fig. 3. While in the former method, antibiotics would loosely reside at the surface of the nanofibers, resulting in an unwanted burst release [114], in the latter one, involving high voltage and high shearing forces imposed at the interface between core and shell fluids, proteins could be rapidly dehydrated and delicate bioactive agents harmed [115]. Emulsion electrospinning and surface coating of the electrospun nanofibers are two other approaches for loading the nanofibers with antibiotics. In the case of emulsion electrospinning, two immiscible aqueous and organic phases containing the antibiotic and the polymer, respectively, are mixed. The resultant emulsion is subsequently electrospun. The as-spun nanofibers could be either consisting of dispersed zones of the inner phase across the outer phase or as core-shell structures made via coalescence of the inner phase, leading to formation of a continuous core. This technique has proved to be efficient for incorporation of proteins within biodegradable polymers. Yet, the likely denaturation of proteins when subjected to organic solvents or under the extreme conditions present within the course of initial emulsification can be a significant shortcoming [22]. Lastly, the antibiotic molecules can be adsorbed onto the nanofibers' surface physically or chemically. This strategy typically needs surface modification of the nanofibers to arise the necessary functionality for the immobilization of the antibiotic molecules. Despite some advantages in terms of immobilization and sustained release of antibiotics, this approach involves several steps of chemical reactions, thus it is complex, and is restricted to nanofiber materials that are stable in aqueous solutions [22]. Table 3 briefly lists a number of state of the art antibiotic loaded biopolymeric nanofibers suggested for wound dressing application.

Despite a large diversity and number of developed antibiotics, due to toxicity effects and challenging cellular uptake, very few of them have been clinically utilized. To date, the only antibiotics that have found application in relation to wound dressings are aminoglycosides, beta-lactams, glycopeptides, quinolones, sulphonamides and tetracyclines [21]. Such drugs fatally affect the bacterial activity through four main mechanisms depicted in Fig. 4a, including disruption of bacterial cell wall biosynthesis, nucleic acid and protein synthesis, and also obstruction of major metabolic pathways [21]. As an example for the antibiotic releasing nanofibrous wound dressings, CS nanofibers have been surface decorated with gentamicin-loaded liposome [116]. To do so, the surface of the CS nanofibers was first thiolated to enable covalent bonding with liposome maleimide groups, Fig. 4b. Such a hybrid system could show a promising bactericidal activity against *E. coli, P. aeruginosa* and *S. aureus*. However, the production cycle of the CS nanofibers through electrospinning necessitates involvement of nonbiocompatible solvents such as trifluoroacetic acid (TFA) that can endanger the health of the adjacent tissue [98]. In another study, Ceftazidime was incorporated into the SF/GE nanofibers [123]. The as-developed natural nanofibrous wound dressing is able to inhibit growth of *P. aeruginosa*, which is the bacterium causing infection in over 50% of burn related wounds. Rath et al. [117] synthesized GE nanofibers containing ZnO nanoparticles and cefazolin for the purpose of accelerating the wound healing process and prevent-

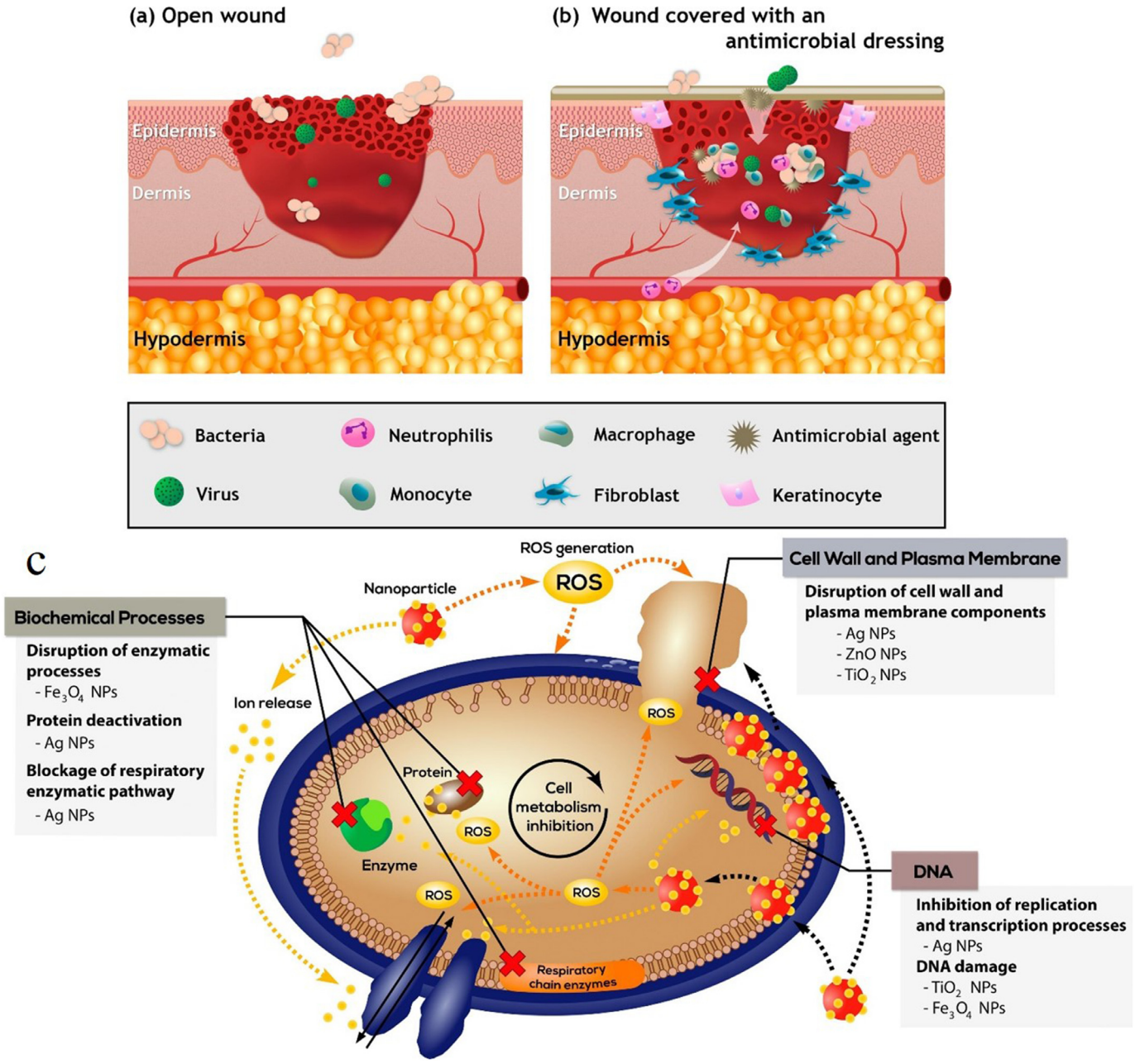

Fig. 2. Schematic illustration of the healing trend in: a) an open wound alone and b) an open wound treated with an antimicrobial wound dressing. As seen here, the open wound is susceptible to invasion of bacteria, engendering delayed healing due to prolonged inflammation and large release of metalloproteinases, potentially destroying the ECM constituents and hindering the generation of new granulation tissue. In case of applying an antimicrobial dressing, the pathway of pathogens into the wound is blocked and the ones penetrated are efficiently killed. Moreover, provoking the immune system and fibroblast/keratinocyte migration, the healing pace is accelerated. c) Various mechanisms suggested for the killing function of antibacterial nanoparticles. Reproduced with permission [21]. Copyright 2018, Elsevier.

ing infection concurrently. Cefazolin acts as an inhibitor against the synthesis of the bacterial cell wall through attachment to specific penicillin-binding proteins and thus confers the wound dressing made of related encapsulating nanofibers with promising antibacterial features. Moreover, ZnO nanoparticles steadily release Zn cations that can largely raise re-epithelialization, lower inflammation, and inhibit bacterial growth. Being a semiconductor, reactive oxygen species (ROS) are generated by ZnO nanoparticles, thereby optimizing cell adhesion, proliferation, and migration via the growth factor mediated pathways. Zn as the cofactor of metaloprotein is vital for the regeneration of the ECM. It also performs as a regulator in the process of auto debridement and keratinocyte migration, two important prerequisites for wound healing. As mentioned earlier, ZnO nanoparticles also show bactericidal potential, thanks to their ROS generation ability that imposes oxidative stress on proteins, DNA, and membrane structures [124]. Fig. 4c implies extensive, homogenous formation of collagen in the tissues treated with the biohybrid nanofibers after 14 days, witnessing a notable wound healing effect. Also, reactive cells (in dark color) and fresh fibrous tissues are explicitly observed. Compared to the control group, the hybrid nanofibers induce formation of a larger number of reactive cells (initial phase) and also provoke a higher fibroblast content.

In the case of the antibiotic loaded nanofibrous dressings, structural properties including nanofiber diameter play a decisive role in the release of the antibiotic molecules. The swelling degree of

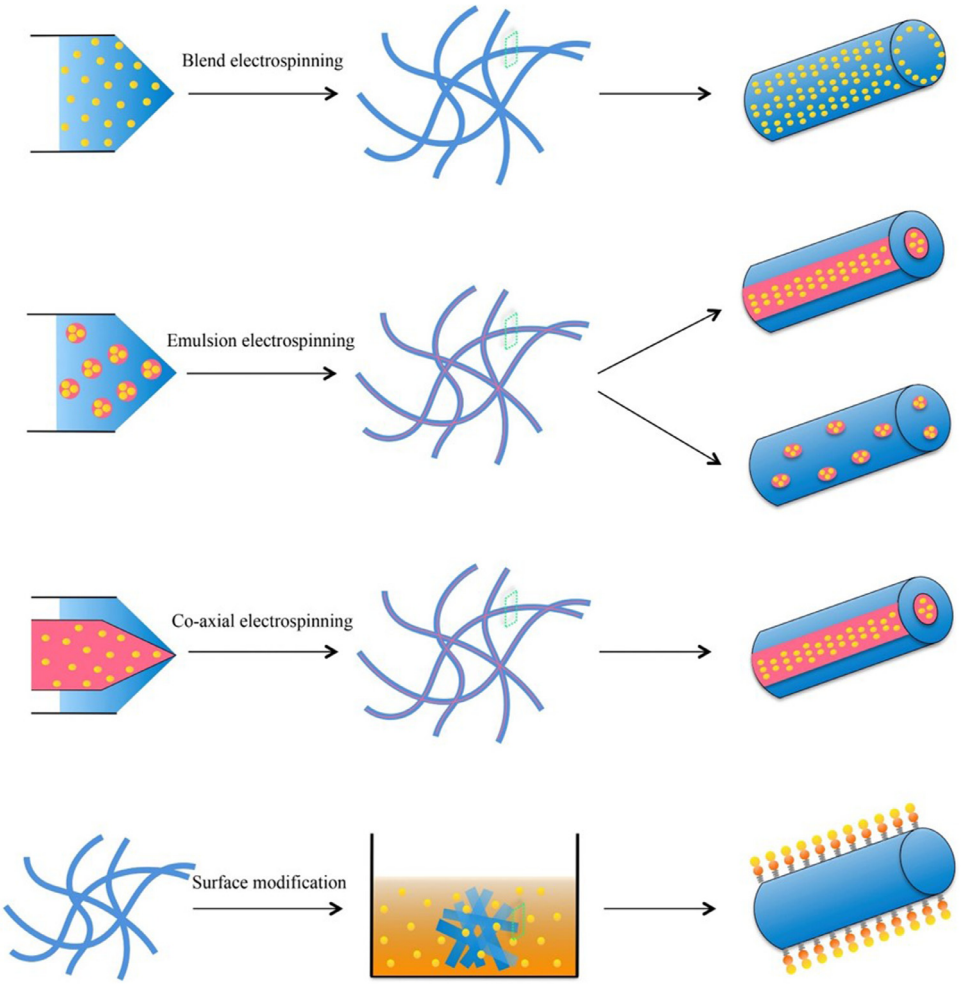

Fig. 3. Various adopted methods for inclusion of antibiotics into nanofibers. From top to bottom: Blend electrospinning based on co-dissolution of antibiotics and polymers in a common solvent; Emulsion electrospinning, based on emulsification of antibiotic solution into an immiscible polymer solution; Co-axial electrospinning, based on separate electrospinning of antibiotic and polymer solutions by two concentric nozzles; Surface decoration of antibiotic molecules on the nanofibers' surface physically or chemically. Reproduced with permission [22]. Copyright 2017, Elsevier.

the nanofibers is dependent on the nanofiber diameter and affects the release kinetics. The finer the nanofibers, the higher the swelling percentage is [117].

Moxifloxacin hydrochloride, that is a fourth-generation quinolone antibiotic, is commonly employed for respiratory infections, community-acquired pneumonia, as well as the infections occurring in skin and soft tissues. Fu et al. [125] included moxifloxacin hydrochloride into blend nanofibers of sodium alginate/PVA to address the bacterial infection in wounds. By releasing around 80% of moxifloxacin hydrochloride after 10 h incubation at 37 °C, the nanofiber wound dressing enables removal of *P. aeruginosa* and *S. aureus*. Alginate fibers have also been used as a carrier for sulfanilamide [126]. This compound is regarded as an inexpensive antibacterial and anti-inflammatory agent, hampering enzymatic reactions based on para-aminobenzoic acid. For such hybrid fibrous wound dressings, a sustained and controlled drug release was recorded, leading to a bactericidal efficiency of 86.4% and 94.6% for *S. aureus* and *E. coli*, respectively.

A wound dressing composed of nonmulberry SF nanofibers functionalized with EGF and ciprofloxacin HCl (CIP), has shown desired properties such as biocompatibility, remarkable water uptake capacity of 440%, optimum water vapor permeability (2330 g m^{-2} day^{-1}), excellent mechanical properties ($E = 2.5$ MPa) and also bactericidal effect [43]. In this system, while EGF provides the necessary growth factor for wound microenvironment, CIP offers an antibacterial effect. In fact, this system enjoys biomimetic and physicomimetic materials that provoke cell-matter interactions and thereby tailor deposition of ECM and healing rate of the wound. The nonhomogenous size distribution of the nanofibers allows for biomimicry of the natural ECM, whose collagen fibrils size varies from 10 to 300 nm [127]. Additionally, cellular activities including migration, proliferation, and differentiation largely depend on the mechanical properties of the dressing. The cell-matter interactions can be tuned by the external shear stress as well as the mechanical signaling channels that govern the cellular behaviors [127]. Hydrophilicity and water uptake capacity are other important factors that can raise the wound healing efficiency. This outcome is achieved through a short re-epithelialization time and proper ECM deposition, thanks to optimized cellular migration within a moist microenvironment [128]. Also, by absorption of wound exudates and rehydration of the devitalized part of wound tissue, moist dressings enable wound autolytic debridement [128]. The architecture of the nanofibrous dressing and its porosity are determining in water vapor permeability. A porous wound dressing with notably large water vapor transmission rate (WVTR) (~5000 g m^{-2} day^{-1}) promptly desiccates the wound milieu and renders it necrotic. In contrast, an impermeable, occlusive dressing shows a negligible WVTR (~300 g m^{-2} day^{-1}), main-

Table 3
Antibiotic loaded biopolymeric nanofibers developed since 2015.

Antibiotic	Nano-fibrous carrier	Antibiotic loading method	Antibiotic loading efficiency (%)	Target microbe	Antibacterial efficiency (ZoI (mm) or%)	Cell Viability (*in vitro*)	Ref
CIP	PVA-SF	Surface coating of the nanofibers	Not reported	*S. aureus, S. epidermidis, E. coli, P. aeruginosa*	Promising antibacterial activity against all four tested bacteria	Significantly high HDF and HaCaT proliferation	[43]
Gentamicin	CS	Surface coating of the nanofibers	17%	*S. aureus, E. coli, P. aeruginosa*	*E. coli* (22 mm), *S. aureus* (23 mm) and *P. aeruginosa* (16 mm)	–	[116]
Cefazolin	GE	Blend electrospinning	93.7 ± 1.4%	*S. aureus*	3.4 cm	–	[117]
TCH	PCL-CA-dextran	Blend electrospinning	1 wt% polymer nanofibers	*S. aureus, E. coli*	Promising antibacterial activity against both tested bacteria	over 160% for fibroblast cells after 6 days	[118]
TCH	PVA/CS	Blend electrospinning	5 wt.% polymer nanofibers	*S. aureus, E. coli*, and *S. epidermidis*	8.8 ± 0.4 mm for *E. coli*, 15.6 ± 0.3 mm for *S. epidermidis* and 19.6 ± 0.2 mm for *S. aureus*	90% after 3 days for SMCs	[42]
Tigecycline	sericin/PVA	Blend electrospinning	84.25%	*E. coli*, and *Bacillus subtilis*	30.13 ± 1.31 for *E. coli* and 40.51 ± 0.65 for *Bacillus subtilis*	comparable cell (L929 fibroblast cell) activity with control	[119]
TFH	PCL/GE	Blend electrospinning	100%	*Trichophyton mentagrophytes, Aspergillus fumigatus* and *Candida albicans*	quite efficient in hindering the growth of *Trichophyton mentagrophytes* and *Aspergillus fumigatus* but unable to suppress that of *Candida albicans*	75% cell (L929) viability after 2 weeks	[120]
CIP	EC	Blend electrospinning	5 and 15 wt.% polymer nanofibers	*S. aureus, E. coli*	4.29 cm for *E. coli* and 4.72 cm for *S. aureus*	100% cell (HDF) viability after 24 h	[121]
Silver sulfadiazine	PVP/GE	Blend electrospinning	0.1, 0.2 and 0.3 wt.% polymer nanofibers	*S. aureus, E. coli*	4 mm for *E. coli* and 3.7 mm for *S. aureus*	–	[122]

*Zone of Inhibition (ZOI); Ciprofloxacin (CIP); polyvinylalcohol (PVA); silk fibroin (SF); human dermal fibroblast (HDF); tetracycline hydrochloride (TCH); terbinafine hydrochloride (TFH); ethyl cellulose (EC); chitosan (CS); gelatin (GE); polycaprolactone (PCL); cellulose acetate (CA); smooth muscle cells (SMCs); polyvinylpyrrolidone (PVP).

tains the exudate and renders the wound vulnerable to infection. In such instances, wound healing is decelerated. An ideal wound dressing should offer a WVTR value of 2000–2500 g m^{-2} day^{-1} [129].

Despite the antibacterial effect and wound healing potential of antibiotics, their frequent application can lead to bacterial resistance. Over 70% of the bacteria engendering wound infections endure in the presence of a minimum of one antibiotic being used in medicine [21]. Additionally, there are several multidrug resistant bacteria and their number is ascending considerably. This means that bacteria are becoming resistant against many varieties of natural and synthetic antibiotics and thus efficient alternative means must be sought promptly. In this context, biohybrid nanofibers involving antimicrobial agents in the form of nanoparticles, biofunctional ligands, antibacterial ion releasing organic or inorganic materials, and plant-derived compounds are potential alternatives.

5.2. Nanocomposite antibacterial nanofibers

Nanofibrous wound dressings with antibacterial action can be classified as those containing antibacterial nanoparticles (nanocomposites), functional agents (surface functionalized), and bio-derived compounds (bioblends). Regarding the first category, silver nanoparticles have endowed polymeric nanofibers with an antibacterial effect [130].

5.2.1. Silver nanoparticle containing nanocomposite antibacterial nanofibers

Silver is considered an important antibacterial agent for infections occurring in burns, open wounds, and chronic ulcers. Its recognition for this purpose (bactericidal activity) has a long history exceeding centuries. For instance, as old as 1000 BC, silver was used for water treatment due to its bactericidal effect [131,132]. Also, over many centuries, Ag has been implemented in the treatment of various diseases [133]. Tetanus and rheumatism are two medical problems that were treated by silver in the 19th century. Also cold and gonorrhea were cured by silver based antibacterial drugs in the early 20th century [131]. Silver is almost an inert metal that is weakly absorbed by mammalian or bacterial cells. Yet, when exposed to wound fluids, it is easily ionized and turns to an extremely reactive agent that sticks to proteins and cell membranes [134]. Silver ion kills microorganisms and drastically damages them by poisoning the respiratory enzymes and the microbial electron carrying sections and also by disabling various DNA activities [134]. The hindering performance of silver ion stems from its intensive bonding ability with thiol groups available in the cell respiratory enzymes. Moreover, silver ion is able to interact with the proteins constituting the cell structure and particularly with DNA bases, thereby hampering replication [132]. According to the *in vitro* studies, the bactericidal effect of the silver ion is mainly ascribed to its attachment to the unoccupied sulphydryl groups within the bacterium or on its surface, engendering

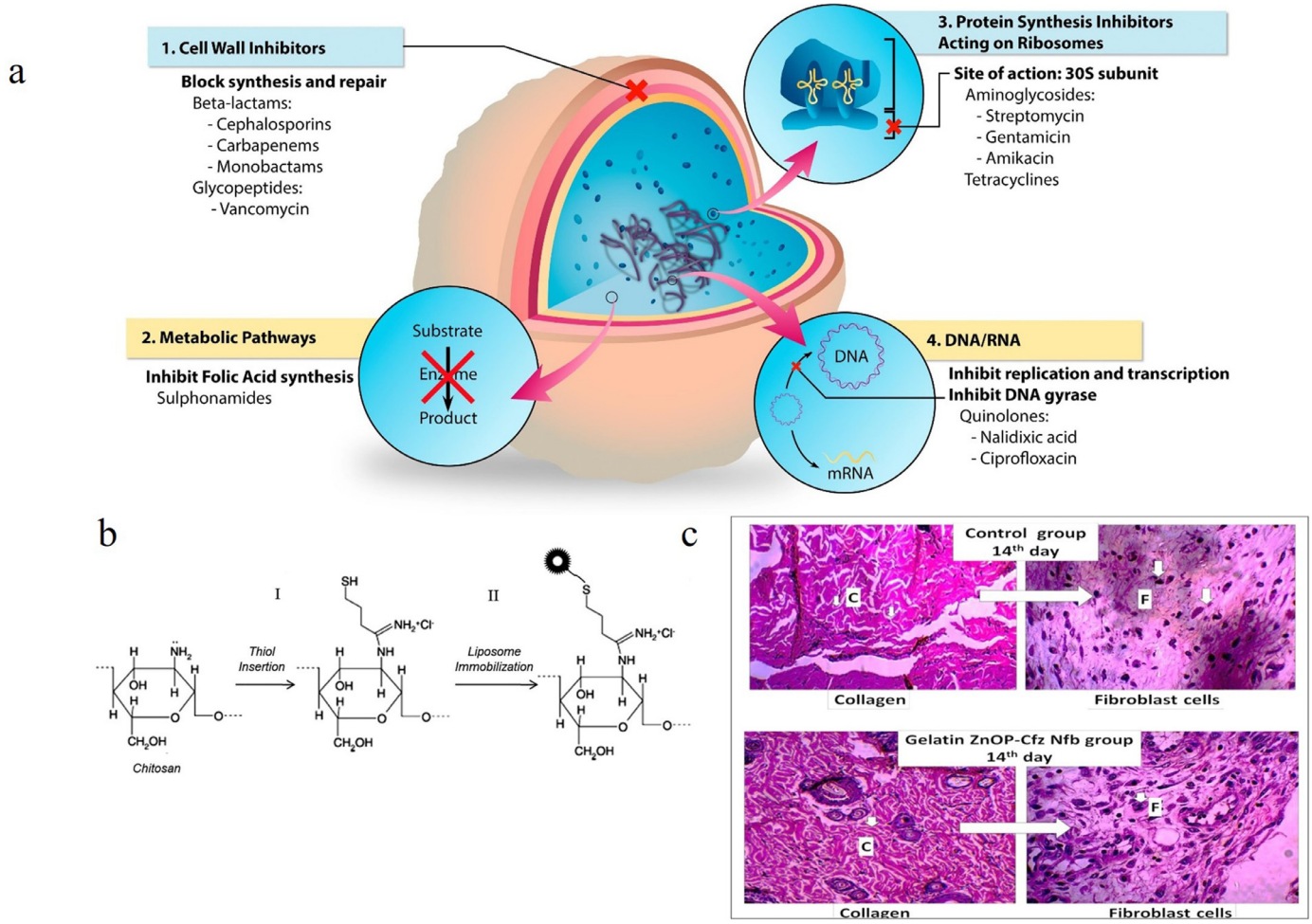

Fig. 4. a) The four possible mechanisms whereby antibiotics disrupt the bacterial activities. Reproduced with permission [21]. Copyright 2018, Elsevier. b) The chemistry behind the liposome immobilization on the chitosan nanofibers' surface comprising: I – the reaction between the primary amine group of chitosan and 2-iminothiolane (2-IT) at pH 7.27; II – emergence of surface thiol groups and their reaction with maleimide at pH 7. Reproduced with permission [116]. Copyright 2015, Elsevier. c) H&E staining of the tissue adjacent the control and cefazolin–ZnO nanoparticle incorporated gelatin nanofiber implies the collagen content (C) and density of the fibroblast cells (F). Reproduced with permission [117]. Copyright 2016, Elsevier.

passivation of the enzyme phosphomannose isomerase [135]. Additionally, accumulation of silver ion within the cell and its likely interaction with the cytosolic proteins, mitochondrial enzymes and nuclear DNA or RNA complement this bactericidal function [131,135]. Fig. 2c demonstrates the underlying mechanisms for the bactericidal effect of Ag nanoparticles.

Given the high potential of silver ion in addressing the significant challenge of wound infections, Ag nanoparticles have been widely employed as an antibacterial filler for polymeric nanofibers developed for wound dressings. In general, incorporation of Ag nanoparticles into polymeric nanofibers can be carried out in several manners. One approach is electrospinning of a dispersion of Ag nanoparticles in a polymer solution. Such method has been already shown to be problematic due to aggregation leading to the loss of efficiency of the nanoparticles [14,15]. In contrast, formation of Ag nanoparticles onto the polymeric nanofibers seems a favorable technique in terms of preservation of the antibacterial activity of the nanoparticles. In this regard, several techniques have shown promising applicability including sol-gel and surface functionalization. As the former method, inclusion of a precursor salt to the polymer solution to be electrospun and eventually hydrothermal treatment of the formed nanocomposite nanofibers, bring about homogenous surface decoration of the nanofibers by the nanoparticles [15,136,137]. Surface functionalization by proteins, polydopamine, among others, could be also an effective approach to create polymeric nanofibers decorated by metal nanoparticles [13]. For instance, an inexpensive serum albumin protein such as Bovine Serum Albumin (BSA) can readily bond with the functional polymers, e.g. poly(acrylonitrile-*co*-glycidyl methacrylate) (PANGMA), through an amine-epoxy reaction. This biofunctionalized nanofiber system can subsequently capture noble metal nanoparticles through a metal-protein interaction when immersed in a metal containing aqueous dispersion. Under this condition, the protein undergoes conformational change from alpha helix to beta sheet (Fig. 5a) and thereby exposes functional groups that can capture metal nanoparticles, biomolecules, etc. (Fig. 5b) [13]. The resultant nanocomposite nanofibers thus exhibit nanoparticles uniformly coated on the surface, Fig. 5c. Inspired by mussel adhesion onto diverse surfaces in nature, Fig. 5d, polydopamine synthesized through self-polymerization of dopamine under alkaline conditions, has also offered interesting opportunities for surface functionalization [138]. This coating material is able to reduce metal cations through its catechol groups and form metal nanoparticles, e. g. Ag. The as-synthesized Ag nanoparticles are insensitive to oxidation, enabling their longstanding antimicrobial function [130]. One significant challenge related to polydopamine based coatings is the difficult control of the coating thickness and surface morphology. This shortcoming hinders the even distribu-

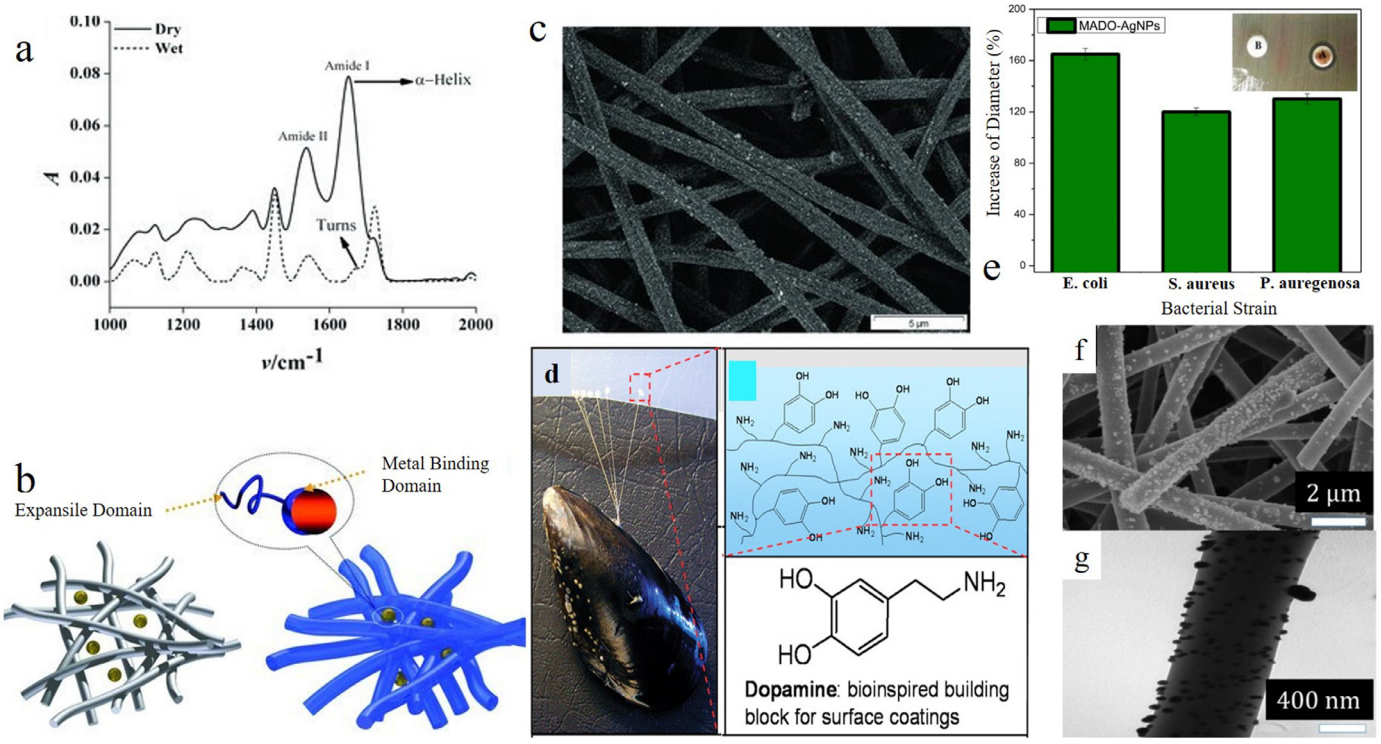

Fig. 5. a) ATR-FTIR spectra for the BSA functionalized PANGMA nanofibers in different hydration states (of dry and wet represented by the solid and dashed lines, respectively). b) Schematic illustration of the capturing process of the metal nanoparticle by the swollen functionalized nanofibers). c) SEM image shows uniform distribution of metal nanoparticles across the nanofibers. Reproduced with permission [13]. Copyright 2012, Wiley-VCH. d) Camera image of a mussel stuck onto a polymeric surface along with a simple representation of the amine and catechol groups of the dopamine building block for surface coating. Reproduced with permission [138]. Copyright 2007, Science AAAS. e) Antibacterial activity of Ag nanoparticle/MADO nanofibers against *P. aeruginosa, S. aureus*, and *E. coli*. The inset image compares the bactericidal activity of the nanocomposite nanofibers with that of the neat ones on a LB-agar plate containing *P. aeruginosa* f) FESEM and g) HRTEM images of the Ag nanoparticle/MADO nanofibers. Reproduced with permission [130]. Copyright 2015, American Chemical Society.

tion of the Ag nanoparticles due to raised surface roughness induced by undesirable aggregation of polydopamine in arbitrary locations [139]. To address this concern, a mussel-inspired copolymer, called poly(dopamine methacrylamide-*co*-methyl methacrylate) (MADO) has been electrospun to develop nanofibers. Such catecholic nanofibers are able to reduce and therefore form Ag nanoparticles on the nanofiber surface [130]. Accordingly, the nanocomposite nanofibers show promising antibacterial performance against both Gram-negative (*Escherichia coli; E. coli*) and Gram-positive bacteria (*S. aureus)* and *Pseudomonas aeruginosa (P. aeruginosa)*), Fig. 5e. Fig. 5f and g exhibit the uniform arrangement of Ag nanoparticles on the surface of the MADO nanofibers. Despite various merits of such nanocomposite strategy, the rather complex material synthesis and likely high costs involved can potentially hamper large scale production of this wound dressing material. Moreover, release and accumulation of silver, that is potentially cytotoxic, in different organs could be problematic and could induce undesirable side effects. For instance, Ag nanoparticles decline the activity of mitochondrial respiratory chain complexes I, II, III, and IV within different tissues of Wistar rats [140]. With respect to Ag nanoparticle incorporated biopolymeric nanofibers for wound dressing purposes, a recent study showed the use of jelly fish as a Q-mucin glycoprotein and collagen rich component blended with polycaprolactone (PCL) in a nanofibrous system [141]. Fig. 6a shows the formation process of the core (PCL)-shell (jelly fish) fibers during electrospinning. Superior to mammalian (e.g. bovine) derived collagen that potentially endangers the health of recipients via transmission of bovine spongiform encephalopathy and transmissible spongiform encephalopathy, collagens obtained from sea creatures such as jelly fish are safe and offer different merits. Other than collagen type I and II, the jelly fish tissue contains Q-mucin glycoprotein. This largely glycosylated high molecular weight nonglobular protein possesses similar structural characteristics to human mucins. As a notable feature, having cysteine amino acid, mucin proteins are able to capture MMPs and neutrophile elastase, which is the collagen-digesting enzyme, thereby facilitating collagen synthesis and faster healing of wounds. To benefit from the promising properties of the jelly fish protein for the sake of wound healing and concurrently addressing possible infections, Nudelman et al. [141] developed a blend nanofiber system comprising jelly fish biomass, qmucin and PCL. This bioblend nanofiber is able to chelate Ag ions through mucin cysteine amino acids whose electronegativity is notably lower in comparison to that of Ag^+, engendering formation of a biohybrid nanofiber with antibacterial activity. Interestingly, growth of *E. coli*, nonpathogenic *S. aureus*, and methicillin-resistant *S. aureus* was totally inhibited by the biohybrid nanofibers, Fig. 6b–d. Also, as shown in Fig. 6e and f, witnessed by preclinical tests based on porcine wound healing models, a prompt and efficient healing trend is achieved using such state of the art biodegradable nanofibers. It was reported that the biohybrid nanofibers can induce wound epithelialization and re-epithelialization and later formation of totally healed skin tissue evidenced by the appearance of a fine and integrated epidermal layer and new collagen fibers in the dermis layer [141]. The authors justify such a healing behavior induced by the biohybrid wound dressing as: 1) there is a close, robust interface between the nanofibers, whose large surface area is coated with collagen and mucin proteins, and the wound. Such an intimate interface notably keeps away MMPs from regenerated collagen and thereby provokes the formation of collagen fibers in the dermis layer. 2) Due to the presence of the Ag nanoparticles, infection is hampered. 3) The ECM biomimicking structure of the jelly fish nanofibrous

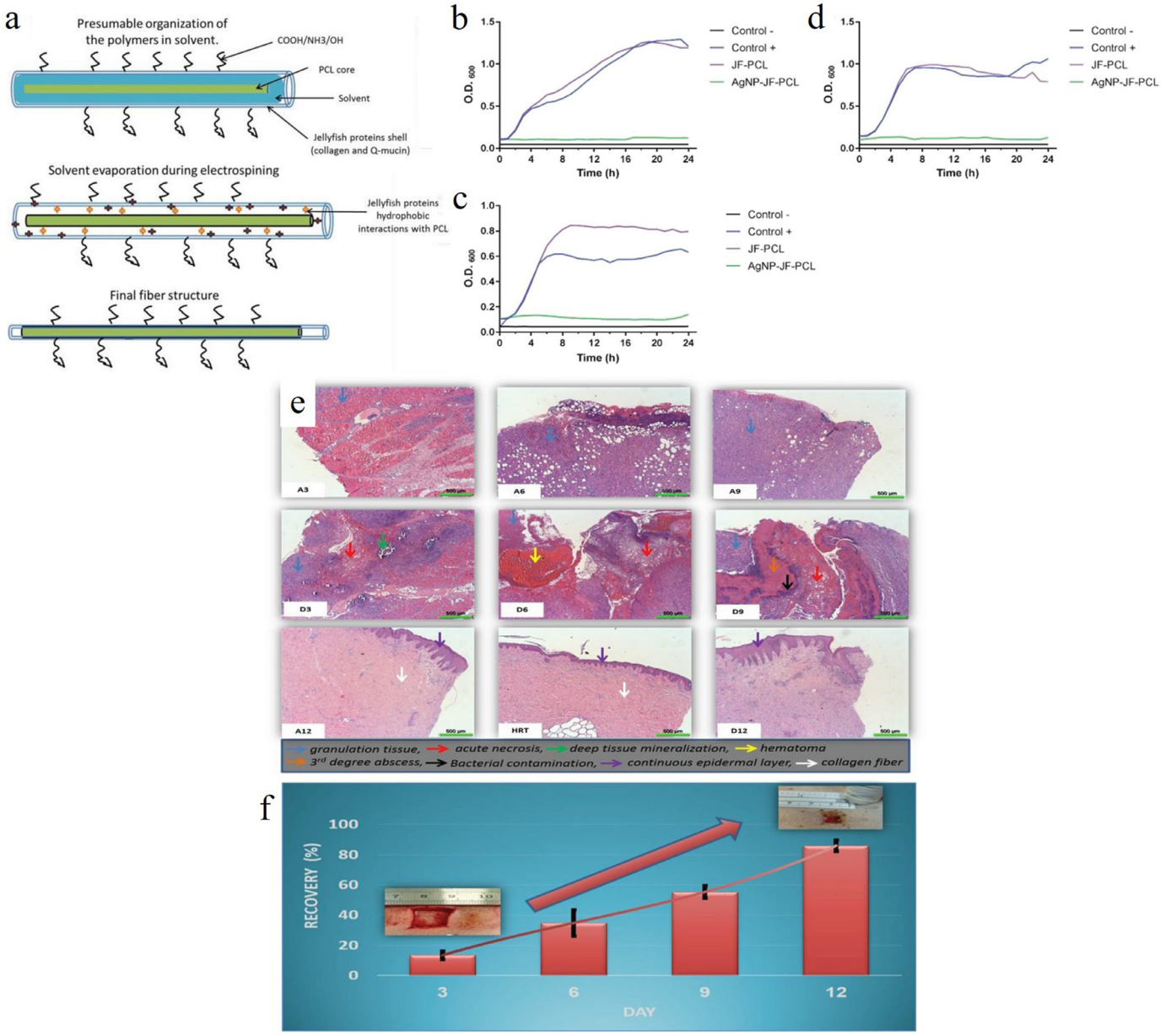

Fig. 6. a) Synthesis procedure of jelly fish (JF)-PCL fibers. b–d) The graphs imply the bacterial growth trends. Note that the negative control is the sterile Tryptic soy (TS) broth alone and the positive control is the TS broth inoculated with b) *E. coli*, c) *S. aureus*, and d) Methicillin-resistant *S. aureus*. e) H&E staining of the wound tissue subjected to the jelly fish (A) and silvercell commercial wound dressing (D), over a 12 day period with intervals of 3, 6, and 9 days. Note that the arrows in different colors represent different processes taking place within the wound. f) The jelly fish dressings raises the recovery percentage of the wounds treated by them. Reproduced with permission [141]. Copyright 2019, John Wiley and Sons.

dressing encourages cellular activities and thus the healing process. 4) Thanks to the hydrophilicity of the nanofibers, moist is maintained and dehydration of the wound and crust formation is avoided.

5.2.2. Metal oxide nanoparticle containing nanocomposite antibacterial nanofibers

As shown in Fig. 2c, alongside Ag nanoparticles, other types of nanoparticles made of Fe_3O_4, TiO_2 and ZnO can potentially act as antimicrobial agents coupled with biopolymeric nanofibers. For instance, Cai et al. [142] developed chitosan/gelatin hybrid nanofibers that were reinforced by inclusion of Fe_3O_4 nanoparticles. This nanocomposite strategy led to formation of nanofibrous dressings with high robustness and antibacterial function. However, agglomeration of the nanoparticles and their residence inside the nanofibers are two bottlenecks that could notably challenge the antibacterial performance of such nanocomposite systems. On the other hand, deposition of the nanoparticles on the nanofiber surface could lead to their release into the human body and having undesired consequences, as they can reach internal sensitive organs and alter cell biochemical pathways.

TiO_2 nanoparticles also offer bactericidal activity when subjected to UV irradiation. This activity can be tailored by changing the irradiation related factors including duration, intensity and wavelength, environmental parameters such as pH and temperature, dimension and morphology of the nanoparticle, oxygenation and hydroxylation degree, and ROS retention time [143,144]. It is worthy to note that the photocatalytic antibacterial activity of TiO_2 nanoparticles can be tuned by doping with metals, metal oxides, or nonmetals [145]. In addition to the light-driven ROS generation, TiO_2 can show antibacterial activity even in the darkness [143]. Outstanding bactericidal activity of TiO_2 nanoparticles has

provoked a number of related researches where this property is coupled with the biocompatibility and the extensive surface area of biopolymer nanofibers. For instance, Woo et al. [146] fabricated a bilayer wound dressing comprising a TiO_2–CS nanofibrous layer overlaid onto a human adipose-derived ECM sheet. In this system, the hydroxyl and glucosidic groups of CS are rich in terms of electron density and can chelate titanium ions, forming a robust bond between the nanoparticles and nanofibers. The as-developed dressing not only inhibits bacterial (*E. coli* and *S. aureus*) growth and infiltration but also shrinks the wound size to 17% in comparison to 23% seen in the controls. The implementation of TiO_2 nanoparticles as an antibacterial additive to polymeric nanofibrous wound dressings has been discussed in the literature [147].

Zn is an important element influencing cellular activity and it is involved in the synthesis of DNA and RNA, thus in association with cell replication, differentiation and transcription. Zn also plays a crucial role in relation to enzyme systems impacting cell division and proliferation [148]. Due to their antimicrobial effect alongside their ability to induce proliferation of fibroblasts and angiogenesis, ZnO nanoparticles have also found applications as nanofiller in nanofibrous wound dressings. Ahmed et al. [79] coupled these nanoparticles with CS/PVA nanofibers to construct a wound dressing for diabetic ulcers that enabled promising antibacterial activity against *E. coli* (20 mm ZoI), *P. aeruginosa* (21.8 mm ZoI), *B. subtilis* (15.5 mm ZoI) and *S. aureus* (21.5 mm ZoI). Moreover, a faster healing rate was recorded for the nanocomposite nanofibers in comparison to the neat control ones. This behavior is attributed to a higher ROS concentration triggered by ZnO nanoparticles, that boosts cell migration and proliferation, thus wound healing. Regarding the antibacterial activity, Chen et al. [149] incorporated ZnO nanoparticles into gelatin nanofibers to create an antibacterial wound dressing. They ascribed the promising antibacterial performance of the nanocomposite nanofibers to the generation of superoxide radicals ($\bullet O_2^-$) by the nanoparticles. Superoxide radicals drastically deteriorate the bacterial cell wall, giving rise to leakage of the cell content, thus a bactericidal effect. It is worthy to note that UV irradiation maximizes the generation of the superoxide radicals, thereby causing a higher level of bacterial death. While being efficient in terms of bactericidal effect, some studies imply also the cytotoxicity of ZnO nanoparticles. For instance, Cho et al. [150] state that among SiO_2, CuO, Co_3O_4 and ZnO nanoparticles, CuO and ZnO are the most toxic ones when tested with respect to *in vivo* acute lung inflammogenicity and *in vitro* cytoxicity.

Quaternary ammonium salts have exhibited extensive antimicrobial performance against many Gram-positive and Gram-negative bacteria, and other microorganisms such as yeast and fungi. In this regard, trimethoxysilylpropyl octadecyldimethyl ammonium chloride (QAS) is a newly developed cationic antibacterial agent whose chemical structure comprises a siloxane group that is bound to quaternary ammonium through a long-chain alkyl [151]. By interacting QAS' siloxane group and polymer's functional groups (e.g. hydroxyl), nanofibrous dressings acquire antibacterial activity. The likely antibacterial mechanism of QAS include: (i) the electrostatic attraction of the quaternary nitrogen with positive charge to the oppositely charged bacterial membrane, engendering the membrane disintegration induced by the unequal surface charge distribution. (ii) insertion of the hydrophobic long-chain alkyl groups of QAS into the bacterial membrane and thereby alteration of its physicochemical property, triggering release of the bacterial content [152]. In a recent study, QAS has been incorporated into PCL/GE nanofibers for wound disinfection purpose [151]. Prior to the GE degradation, the inactivation of bacteria takes place through their interaction with the GE component. Later, when infection occurs, the bacteria release proteolytic enzymes that are able to largely degrade GE. Induced by the matrix' disintegration, QAS is liberated and imposes its bactericidal effect on the available bacteria including *S. aureus* (Gram-positive) and *P. aeruginosa* (Gram-negative). Table 4 summarizes some of the recently developed nanocomposite antibacterial nanofibrous wound dressings.

5.3. Biofunctionalized antibacterial nanofibers

The second class of antibacterial biohybrid wound dressings deals with biopolymeric nanofibers surface functionalized with antimicrobial peptides and amino acids. In this regard, silk fibroin and chitosan are two important biopolymers that enable tethering of antimicrobial agents through their numerous functional groups. As the antimicrobial factor bound to nanofiber surface, AMPs have been widely studied [3]. As mentioned earlier, AMPs have emerged as a new generation of antimicrobial additives to wound dressings given their biocompatibility and, on the other hand, due to the developing bacterial resistance to many antibiotics. In such hybrid systems, the antimicrobial activity of the dressing is tailored by the type of AMP and its activity when immobilized onto the nanofiber surface. The latter factor highlights how crucial the biofunctionalization strategy is. For the sake of AMP immobilization, there are several approaches such as co-spinning, as well as physical and covalent binding. In the case of co-spinning, the possibility of aggregation of the AMP molecules as well as the limited range of proper co-solvents that do not damage the activity of the AMP could be challenging. Regarding physiosorption of the AMPs, the loose linkage between the antimicrobial agent and the nanofiber probably leads to the AMP fast desorption [3]. Among the mentioned approaches, covalent immobilization offers several merits including negligible AMP leaching, long-term stability as well as nontoxicity [158]. There are two main methods for rendering covalent links between AMPs and nanofibers surface, Fig. 7, including: "graft to" and "surface initiated" ones. The former technique is based on the covalent binding of the AMPs onto the nanofiber surface. The latter one deals with polymerization of the AMP from the initiators or spacers containing functional groups that are stabilized on the nanofiber surface through covalent bonds. The "graft to" method involves surface functionalization of the nanofibers via oxidation, UV radiation, atmospheric plasma, ozonation, among others. Such post-treatments allow for emergence of functional groups, e.g. amines, carboxylic acids, aldehydes or thiols [3]. Here, the bottleneck is the steric hindrance which results from the attachment of first molecules, hampering further penetration of the next ones, thus lowering the grafting density [159]. In addition, mechanical stress induced hydrolysis of the bond between the free functional groups and AMPs could occur frequently [160]. Compared to the "graft to" method, the "surface initiated" one enables the peptides to grow at large densities and without steric hindrance. Despite such a merit, the technique is complex and needs precise adjustment of the amount of initiator (e.g. PEG) and substrate (nanofiber) as well as of the radical polymerization parameters. On the whole, advantageous over other relevant approaches, the "surface initiated" one enables a notable controllability of the immobilization process, thanks to direct polymerization of the AMPs on the surface [158]. As a fact, AMP functionalization of the nanofibrous wound dressings is indeed an advanced concept that requires further strict evaluation. Such studies should confirm the long term applicability of these advanced dressing materials in the control of microbial infections without harming the human body's immune system. Given the positive charge of AMP functionalized nanofibers, there is a possibility of adhesion of negatively charged bacteria and likely blockage of pores in the dressing, thus disruption of gas/nutrient exchange pathways can occur. Moreover, since biopolymers *per se* trigger particular biological responses, it is of importance to minimize the likely overlap and inhibition of their effects and those of the AMPs. In the case of functionalization of nanofibrous wound

Table 4
Diverse antibacterial biohybrid nanofibers developed since 2015.

Nanofiber Material	Anti-microbial agent	Synthesis method	Target microbe	Antibacterial efficiency (ZoI* (mm) or%)	Cell Viability (*in vitro*) (%)	Wound healing efficiency (*in vivo*)(%)	Ref
Ag/ MADO	Nano-particle	Electrospinning and Ag reduction via catechol redox chemistry	*P. aeruginosa, S. aureus*, and *E. coli*	*E. coli* (164%), *S. aureus*, (121%), *P. aureginosa*, (133%)	85% *NIH3T3-L1* fibroblast	92%	[130]
Fe_3O_4/CS/ GE	Nano-particle	Blend electrospinning of Fe_3O_4 nanoparticles, chitosan, and gelatin	*E. coli* and *S. aureus*	*E. coli* (21 mm) *S. aureus* (20 mm)	–	–	[142]
TiO_2/CS	Nano-particle	Plasma enhanced chemical vapor deposition of TiO_2 nanoparticle on CS nanofibers	*S. aureus*	11.5 mm	–	–	[153]
Insulin-CS/ PCL-collagen	Nano-particle	Blend electrospinning of PCL/Collagen nanofibers then their dip coating by the CS nanoparticles	–	microbial penetration was characterized based on BHI broth turbidity which was less significant for the nano-composite nanofibers.	as high as the control	63.66 ± 3.03% and 96.90 ± 1.11 at 7 and 14 days after wounding	[154]
ZnO/GE	Nano-particle	Blend electrospinning of ZnO nanoparticles and gelatin	*E. coli* and *S. aureus*	promising antibacterial activity in dark and UV irradiated states	No particular cytotoxicity	–	[149]
GA/CA	Bio-derived	Blend electrospinning	*S. aureus*	*17.5 mm*	–	–	[62]
MH/SF	Bio-derived	Blend electrospinning of SF, PEO and MH	Methicillin-resistant Staphylococcus aureus (MRSA), *P. aeruginosa, E. coli* and *S. aureus*	51% *E. coli* (29% *S. aureus*, 57% *P. aeruginosa*, and 40% MRSA) for MH (70%)/SF	*L929* cells viability prevails that on control	almost 100% after 12 days	[98]
Lawsonia Inermis (henna)/ GE/OST	Bio-derived	Blend electrospinning of henna, GE, and OST	*E. coli* and *S. aureus*	% 92.21 *S. aureus* and 95.13% *E. coli* after 48 h	enhanced fibroblast proliferation with henna addition	73%	[155]
CN, LG and PM/ CA	Bio-derived	Blend electrospinning of cellulose acetate and the essential oils	*E. coli*	100% *E. coli*	*NIH3T3* fibroblasts (after 96 h, 47% for CA/CN, 73% for CA/LG and 75% for CA/PM) and HaCaT keratinocytes (after 96 h, 75% for CA/CN, 90% for CA/LG and 80% for CA/PM)	–	[156]
Lawsone/PCL-GE	Bio-derived	Coaxial electrospinning of PCL (shell polymer) and lawsone-GE blend (core material)	*S. aureus* and *P. aeruginosa*	*S. aureus* (22.43 mm) and *P. aeruginosa* (-)	*HGF cells* (for the nanofibers containing 1.5 wt.% lawsone) *(≈80, 60, and 50% after 1, 2, and 3 days, respectively)*	100% (for the nanofibers containing 1.5 wt.% lawsone)	[157]
Cys-KR12/SF	Bio-functional	SF electrospinning and post treatment by EDC/NHS and via thiol-maleimide click chemistry	*S. aureus, S. epidermidis, E. coli*, and *P. aeruginosa*	99% for *S. aureus*, and *E. coli* at 4 ◦C (at 37 ◦C the *S. aureus* bacterial reduction% declines down to 20% over three weeks)	promoted the proliferation and differentiation of *HaCaT* cells and the proliferation of *NHDF* cells	–	[97]
CH-D/CH-Arg	Bio-functional	binding L-arginine to CH mediated by EDC/NHS that amidates the primary amine groups (glucosamine; GlcN) of CH. The CH-D/ CH-Arg-blend was subsequently electrospun.	*E. coli* and *S. aureus*	≈100% for both the bacteria	100% cell viability for Human fibroblast	75–80%	[80]

* Zone of inhibition (ZoI); cellulose acetate (CA); gallic acid (GA); poly(dopamine methacrylamide-*co*-methyl methacrylate) (MADO); manuka honey (MH); polyethylene oxide (PEO); oxidized starch (OST); cinnamon (CN); lemon-grass (LG); pepper-mint (PM); 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride /N-Hydroxysuccinimide (EDC/NHS); normal human dermal fibroblast (NHDF); chitosan (CH); deacetylated (D); arginine (Arg).

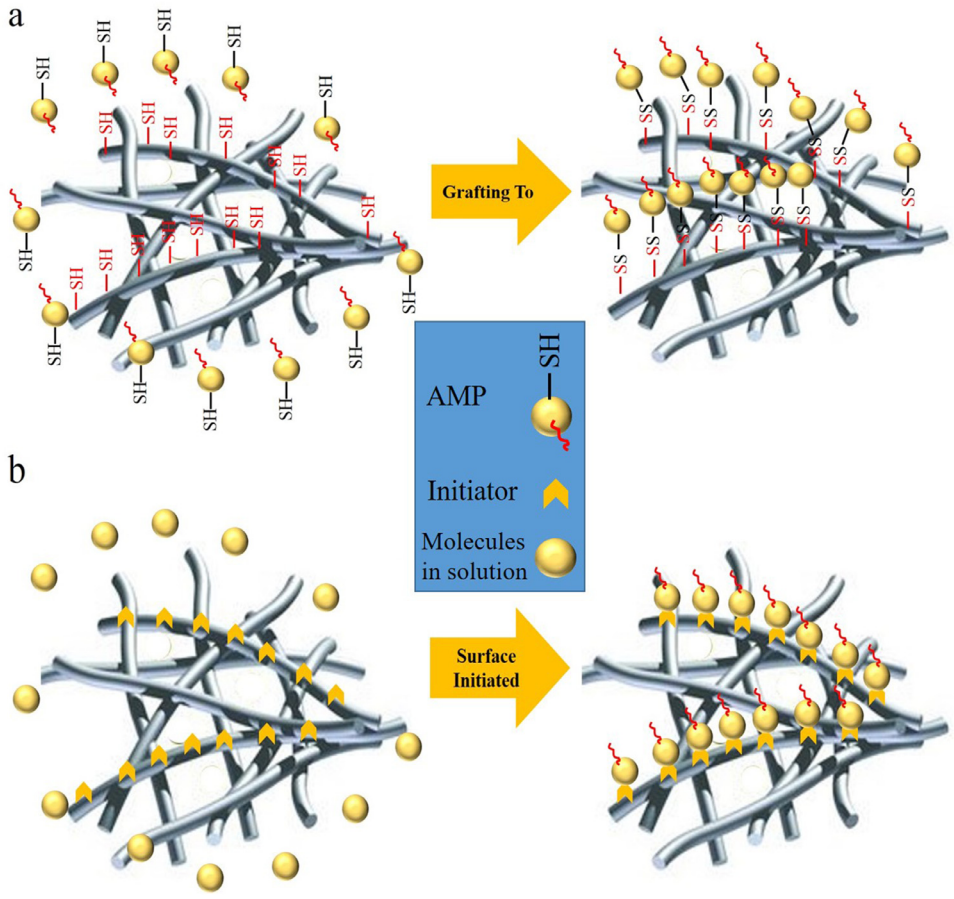

Fig. 7. Different strategies for covalent immobilization of AMPs onto nanofibers' surfaces as: a) "graft to"; and b) "surface initiated" (re-drawn based on the schematic presented in [3]).

dressings with antimicrobial peptides, the interested readers are referred to a previous review article [3].

Given the optimum surface functionality of the biopolymers, the antimicrobial agents can readily be immobilized without any need of post treatment. For instance, there are several antibacterial biohybrid nanofibrous dressings that have been synthesized solely based on surface functionality of SF. Possessing a plethora of functional groups including carboxyl, amine, hydroxyl, and phenol, different antibacterial agents can be readily loaded on SF nanofibers [161]. For instance, antimicrobial peptides (e.g. Cys-KR12 originated from human cathelicidin peptide LL37) have been tethered on SF nanofibers through a thiol-maleimide coupling method, Fig. 8a [97]. The biohybrid SF nanofibers notably reduced bacterial growth when compared with their neat counterparts. Fig. 8b shows that the higher the immobilized factor amount, the larger the antibacterial activity is. The bactericidal effect of the biofunctionalized nanofibers is consistent over a long three week period, regardless of the storing temperature, Fig. 8c and d. FESEM images, Fig. 8e, compare the biofilm formation on two groups of nanofibers, the neat and the biofunctionalized ones, showing that no biofilm appears on the latter group. One developing concern about the cathelcidine peptide is bacterial resistance. For instance, *S. aureus* bacteria are able to reduce the efficiency of such AMPs in different ways. They can lower their surface negative charge when covalently modifying anionic molecules (e.g. teichoic acids) or keep the AMPs away by their pumps or via changing the fluidity of their membrane, and enyzmatic cleavage of the AMPs using proteases [162].

Chitosan is another biopolymer that is biocompatible and biodegradable and offers remarkable antimicrobial effect against a variety of microorganisms including fungi, bacteria, algae, and viruses [163–167]. This performance is achieved via the electrostatic interactions of chitosan amine groups (glucosamines) with positive charge and the counter-charged ones on the microorganism cell wall (e.g. peptidoglycans) [165]. As a result, permeability of the cell wall alters, engendering internal osmotic imbalances. Such mechanism hinders the growth of microorganisms. Also, the hydrolysis of peptidoglycans triggered by the electrostatic interactions expedites the leakage of intracellular electrolytes [21]. Inspired by the promising antimicrobial activity of CS, a variety of blend and functionalized CS nanofibers have been suggested. Given the fact that electrostatic interaction enables chitosan to deactivate and kill bacteria, enhancement of the number and density of the positively charged groups can raise the bactericidal potential of CS. In this regard, positively charged amino acids including L-asparagine [168], L-arginine [169], and L-lysine have been grafted onto CS to enlarge the density of the positive charge. For instance, a wound dressing has been constructed based on deacetylated/arginine functionalized chitosan [80]. Having the guanidine group (pKa = 12.5), at physiological pH, the CS nanofibers functionalized with L-arginine acquire an increased number of positively charged groups, thus show a higher bactericidal effect. Additionally, this biofunctional component confers the dressing higher collagen deposition ability and thereby promotes the wound healing process.

5.4. Bio-derived blend antibacterial nanofibers

Plant-derived compounds such as essential oils and honey are other possible additives for biopolymeric nanofibers. Since pure forms of these compounds are not electrospinnable, they are usually blended with synthetic or natural polymers [170].

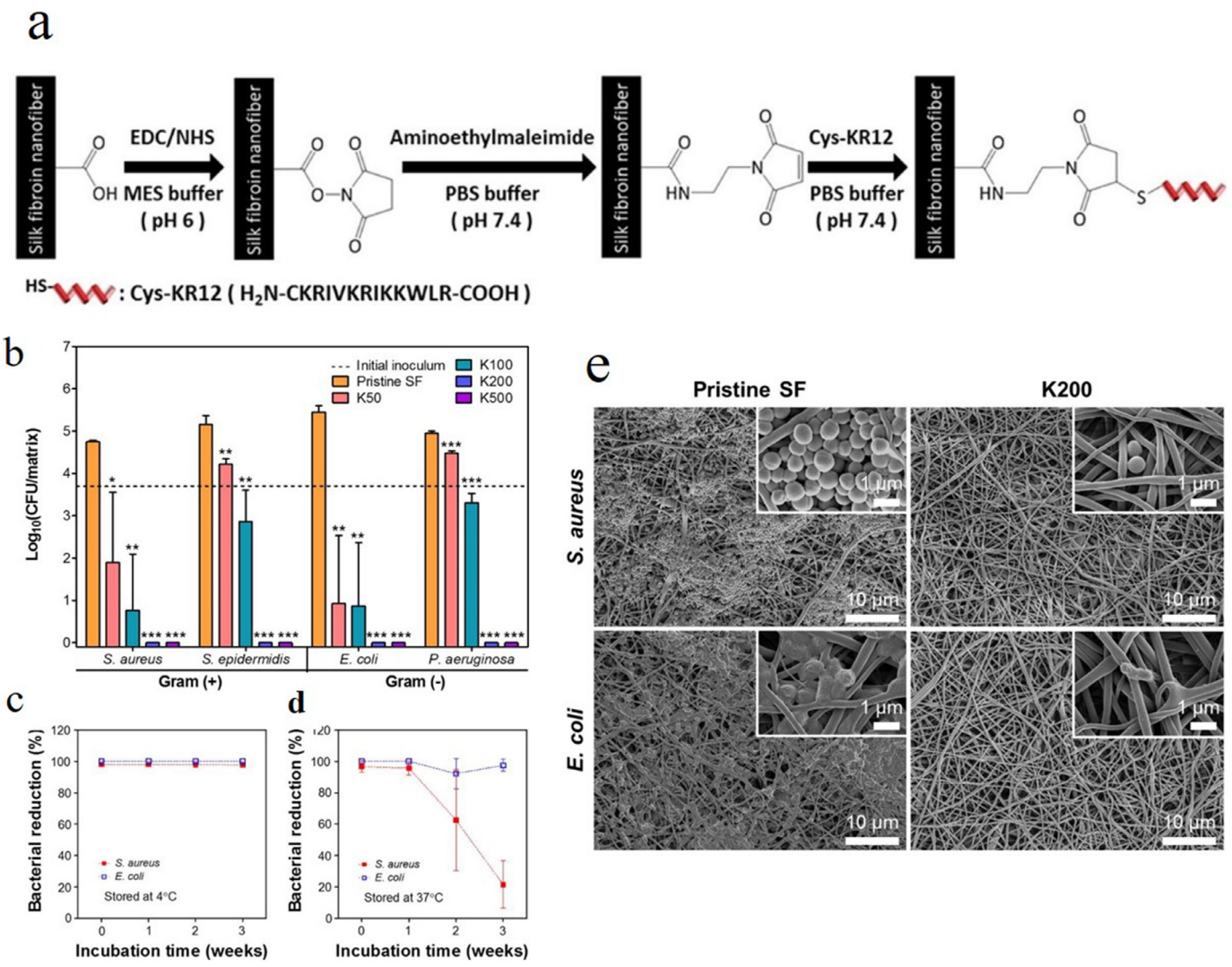

Fig. 8. a) Schematic illustration of the surface functionalization protocol of SF nanofibers by loading of Cys-KR12 mediated by EDC/NHS and via thiol-maleimide click chemistry. b) Antibacterial activity of neat SF nanofibers versus those surface functionalized with Cys-KR12, represented by Log(CFU/matrix) (K50, 100, 200, and 500 denote the different extents and densities of the antibacterial agent immobilized and correspond to 50, 100, 200, and 500 μg/mL Cys-KR12) ($n = 3$, mean ± SD); bactericidal activity of the nanofibers over a three week period at (c) 4 °C and (d) 37 °C. The bacterial reduction (%) was determined versus the CFU of neat SF ($n = 3$, mean ± SD). e) FESEM images show the surface of the nanofibers as neat (left column) and functionalized with Cys-KR12 after 24 h culture of *S. aureus* and *E. coli*. Reproduced with permission [97]. Copyright 2016, Elsevier.

Possessing antiviral, antioxidant, insecticidal, anticancer, anti-allergic, anti-inflammatory, and antimicrobial properties [171], essential oils, that are plant secondary metabolites, have been combined with biopolymeric nanofibers for wound dressing application. The antibacterial effect of essential oils is related to their phenolic compounds, particularly thymol (THY) and carvacrol [172]. Cinnamaldehyde, geraniol, thymol analogues, menthol and carvacrol are typical essential oils that have been frequently employed for their antibacterial effect. For instance, antibacterial cellulose nanofibers have been produced by incorporation of three kinds of essential oils, including cinnamon, lemongrass and peppermint [156]. These biohybrid nanofibers could kill *E. coli*, while they remained biocompatible.

Honey is an ancient (as old as 2000 BCE) therapeutic nutrition, that also shows remarkable antimicrobial, anti-inflammatory and antioxidant performance [173,174]. These features have been so appealing that have justified commercialization of a variety of honey containing wound dressings [175]. Thanks to its acidic nature (low pH of 3.5–4), honey is able to acidify the alkaline medium of chronic wounds, thereby stimulating wound healing. This correlation of wound healing to acidic pH of the wound bed has been almost ignored in the development of nanofibrous wound dressings. Therefore, such aspect needs to be addressed by carrying out research on developing nanofibrous interactive dressings that can acidify the wound milieu and thus provoke wound healing. Manuka honey (MH) is one of eight known (studied) kinds of honey that offers antibacterial function at an expansive concentration domain ranging from 6.25% to 50% (v/v). The bactericidal effect of honey is ascribed to its negligible water content, acidity, and also its antimicrobial agents including antimicrobial peptide bee defensin-1, flavonoids, phenolic acid, and hydrogen peroxide [176]. MH notably kills bacteria even at high density colonies, usually with no cytotoxicity effect on skin cells [177]. On the other hand, honey is able to repair skin through encouraging the release of cytokines and stimulating the immune system response tackling infection [178]. Hybrid MH/SF nanofibers have shown applicability for wound healing purpose [98]. They not only increase cell growth and adhesion but also provide antibacterial activity, thereby accelerating wound healing. Very recently, Tang et al. [87] developed honey loaded nanofibers of PVA/alginate blend for wound dress-

ing applications. By increasing the honey content, the antioxidant effect of the hybrid fibers raised and thus excessive generation of ROS, which is tightly associated with wound inflammation and chronicity, was hampered. The biohybrid dressing was proved efficient in terms of antibacterial function, biocompatibility as well as cell viability. When the honey content was as much as 10%, NIH/3T3 cell viability in the proximity of the biohybrid nanofibers rised to 102.71 ± 1.31%.

As an antibacterial agent, herbal extracts have also attracted wide attention, thanks to their negligible side effects and inexpensiveness in comparison to synthetic drugs. A combination of herbal extracts with biopolymeric nanofibers is indeed promising for wound dressing application. In this context, astragaloside IV loaded-SF-GE nanofibrous mats have been beneficial in terms of cell adhesion and proliferation, raised angiogenesis, and thereby expedited healing process with no scarring [179]. *Astragaloside IV* is a bioactive saponin extracted from the dried plant roots of the genus Astragalus. This extract is employed in traditional Chinese medicine. Lawsonia Inermis L. (called as Henna), a member of the Lythraceae family, has been applied as a herbal therapeutic for many centuries, thanks to its antidiabetic, antioxidant, hepatoprotective, hypoglycemic, and anticancer characteristics. Additionally, due to its anti-inflammatory and antibacterial effects, this herb can be employed for treatment of burns, skin infections, wounds and ulcers [155]. *Lawsone (2–hydroxy-1, 4-naphthoquinone)* is the main coloring and bioactive component of Henna Leaves that is used for the development of many derived therapeutics for wound healing applications [180]. Particularly, it has been reported that such compounds are able to accelerate the wound closure process and to raise skin disintegration strength, tissue granulation, hydroxyproline amount, and proliferation of fibroblasts [181]. Despite these attractive merits, cellular activities including migration and cell-cell communication could be challenged due to the physical form of the henna derivatives. This barrier hinders the long term utility of these compounds for treatment of chronic wounds, as it reduces the healing rate and raises the chance of wound infection. Electrospinning enables to benefit from the promising merits of henna while offering a system that suits the wound milieu adequately [182]. For instance, a state of the art biohybrid nanofibrous dressing has been made from Lawsonia Inermis (henna) incorporated gelatin-oxidized starch nanofibers for the purpose of curing second-degree burn wounds. Inclusion of henna into the gelatin-oxidized starch nanofibers optimized adhesion and proliferation of fibroblasts, collagen secretion, and also antibacterial efficiency [155]. Similarly, henna has been incorporated into poly(l- lactic acid) (PLLA)-GE nanofibers to construct wound dressings [183]. A nanofibrous dressing comprising henna embedded CS nanofibers was developed by Yousefi et al. [180]. The biohybrid nanofibrous dressing could show a bactericidal effect against both Gram-positive (*S. aureus*) and Gram-negative bacteria (*E. coli*) to maintain high cell viability and to accelerate wound healing. The bactericidal effect of henna is ascribed to its exposed hydroxyl groups. Such functionality allows for interaction of the henna derivative with the proteins and carbohydrates composing the cellular wall of bacteria, thereby blocking the enzyme sites and inactivating them [184]. An improved cellular (Normal Human Fibroblast foreskin; *NHF*) activity in terms of adhesion and proliferation was also recorded on the nanofibrous dressing containing the henna extract. Eventually, the wound area in the presence of henna containing nanofibers declined down to 3%, confirming the significant impact of the dressing on the wound healing process.

THY, as aforementioned, is an important phenolic compound of the essential oils that can be derived from Lippia gracilis. With various beneficial antioxidant, antimicrobial, anti-inflammatory, and antinociceptive properties, it is regarded a useful additive to biopolymeric nanofibers for the purpose of wound dressing. For instance, recently, Miguel et al. [185] developed a bilayer asymmetric membrane comprising SF/PCL nanofibers and THY-SF/HA nanofibers simulating the skin epidermis and dermis layers, respectively. The as-developed membrane was optimally porous, robust, and wettable. Moreover, it encouraged fibroblast cells to adhere and proliferate onto the structure. Fig. 9a shows that there was no particular cytotoxic effect on fibroblast cells by the nanofibers, including those containing THY. This finding was further verified by the determination of the cellular DNA content, Fig. 9b. As shown in the SEM images, Fig. 9c, compared to the control, cells favorably adhered onto the nanofiber membranes whose topography and roughness resembled the ECM, as reflected in their increased number of filopodia protrusions. The promising viability of fibroblasts on the nanofibers is also explicitly visible on CLSM images, Fig. 9d. Inclusion of THY conferred the membrane antibacterial activity against *S. aureus* and *P. aeruginosa*. While THY-SF/HA nanofibrous membranes reduced the bacterial growth as much as 87.42% and 58.43% for *S. aureus* and *P. aeruginosa*, respectively, the THY free nanofibrous ones reduced this activity for as low as 4.05% and 3.42%, respectively, Fig. 9e and f. Interestingly, as confirmed by SEM images, no significant biofilm formation was recorded on the samples incorporating THY, Fig. 9g.

Starch is a plant-derived carbohydrate compound composed of amylose (20–30%) and amylopectin (70–80%). Diverse merits of this biopolymer are biocompatibility, biodegradability, nontoxicity, large availability and inexpensiveness, enabling large applicability of starch for various utilities in biomedicine and wound dressing [186]. Blending of starch with other biopolymers such as chitosan for wound healing purpose has been studied [78]. Starch is assumed to confer chitosan nanofibers optimized bioactivity as well as water absorption potential. A further addition of PVA into such a blend can raise mechanical stability of the system via inter- and intra-molecular hydrogen bonding of PVA and chitosan side chains. Accordingly, biohybrid wound dressings are able to withstand against the exerted stresses during wound healing in both dry and wet states. Thanks to the co-existence of chitosan, the amazing bactericidal effect of the dressing was confirmed in the presence of *E. coli* (47–72%) and *S. aureus* (60–84%) bacteria. Furthermore, L929 fibroblast cell viability for the nanofibrous dressing was recorded to be as much as 68–98% after 48 h co-culturing, witnessing promising cytocompatibility of the biohybrids. This feature was further validated by a scratch assay, showing major coverage of the wound gap area induced by cell growth and migration. Despite the desirable *in vitro* results, results of *in vivo* testing are missing for such a system. Moreover, the role of starch in the enhancement of biological as well as mechanical performance of the dressing has not been highlighted. The raised mechanical properties and biological activities including cell viability and antibacterial efficiency are attributed to the presence of PVA and chitosan, respectively.

Carica papaya is a common natural drug for treatment of diverse medical conditions including neurological, vascular, and dermal diseases. A notable wound healing, minimized infection potential and low odor generation have been reported for the wounds treated with aqueous derivative of the *Carica papaya* leaves [187]. These leaves largely contain vitamin C (Ascorbic acid), that is a cofactor for lysine and proline hydroxylases, playing a vital role in the formation and endurance of collagen tertiary structure. Moreover, induced by the composition of *Carica papaya*, the genes enabling collagen formation are readily expressed [188]. Fig. 10a shows the underlying mechanism for wo und healing induced by the composition (particularly vitamin C) of the *Carica papaya* leaves. Very recently, Ahlawat et al. [189] investigated the extract of *Carica papaya* leaves in wound healing and blended within PVA/GE nanofibers. This biohybrid dressing showed remarkable antibacterial performance against *E. coli* and *S. aureus* so that a larger in-

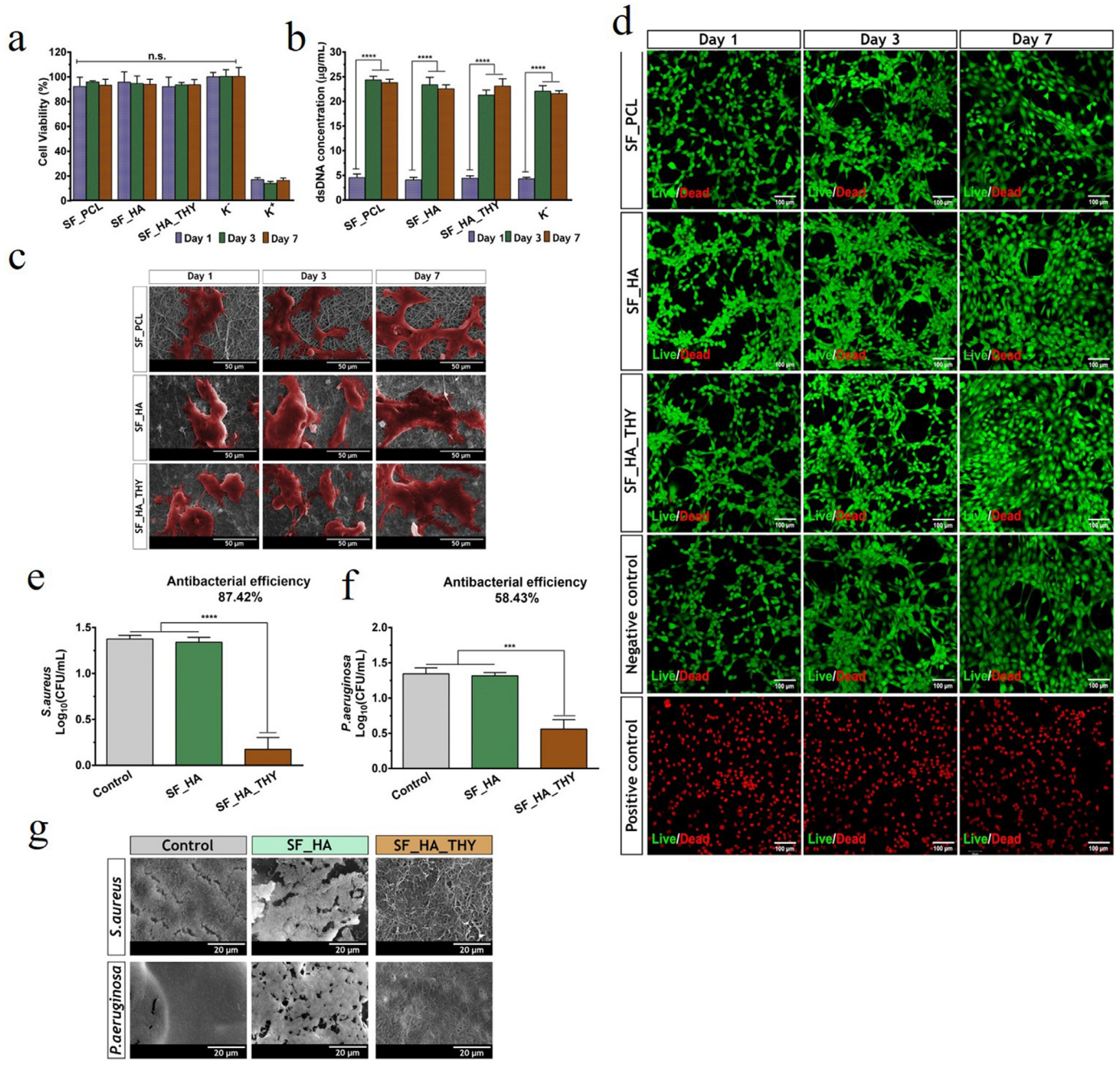

Fig. 9. *In vitro* biocompatibility of THY-SF/HA nanofibers represented by NHDF cell viability (a) and dsDNA content (b) (number of measurements; n = 5, ****p < 0.0001), c) SEM images show the morphology of NHDF cells co-cultured with the nanofibrous samples over a 7 day period. d) Live and dead NHDF cells on the nanofibrous membranes represented by green (Calcein stained) and red (Propidium iodine stained) spots, respectively, in the fluorescence microscopic images. Antibacterial efficiency of the THY-SF/HA nanofibrous membranes against *S. aureus* (e) and *P. aeruginosa* (f) (number of measurements; $n = 5$, $^{***}p < 0.001$, $^{****}p < 0.0001$). g) SEM images show the extent of antibacterial activity of the THY-SF/HA nanofibers in comparisonn to that of the SF/HA nanofibers and the control. Reproduced with permission [185]. Copyright 2019, Elsevier.

hibition zone is formed on the agar plate containing the *Carica papaya*/PVA/GE nanofibers compared to that with the nanofibers free of *Carica papaya,* Fig. 10b–j. Also, as shown in Fig. 10k, the biohybrid nanofibers enabled 80% NIH 3T3 fibroblast cells live adjacent to the nanofibers after 24 h, implying their favorable biocompatibility. Moreover, no particular change in cell morphology was recorded for the cells co-cultured with the biohybrid nanofibers as compared to the ones present in the control medium. Fig. 10l–n compares the seeded cells' morphology on the *Carica papaya*/PVA/GE nanofibers with that of the cells subjected to the PVA/GE nanofibers and the nanofiber free medium after staining with Hoechst 33,342-Rhodamine B. Table 4 summarizes some of the recently developed bio-derived blend antibacterial nanofibrous wound dressings.

6. Conclusion and outlook

Electrospun nanofibers have shown promising potential for a diverse range of advanced applications in environmental remediation, energy, and biomedicine. This attractive potential stems from a number of advantages including interconnected tunable porosity, biomimicry, and large specific surface area enabling engineering of the surface to include a plethora of functional groups as well as immobilization of drug molecules. These features have drawn the

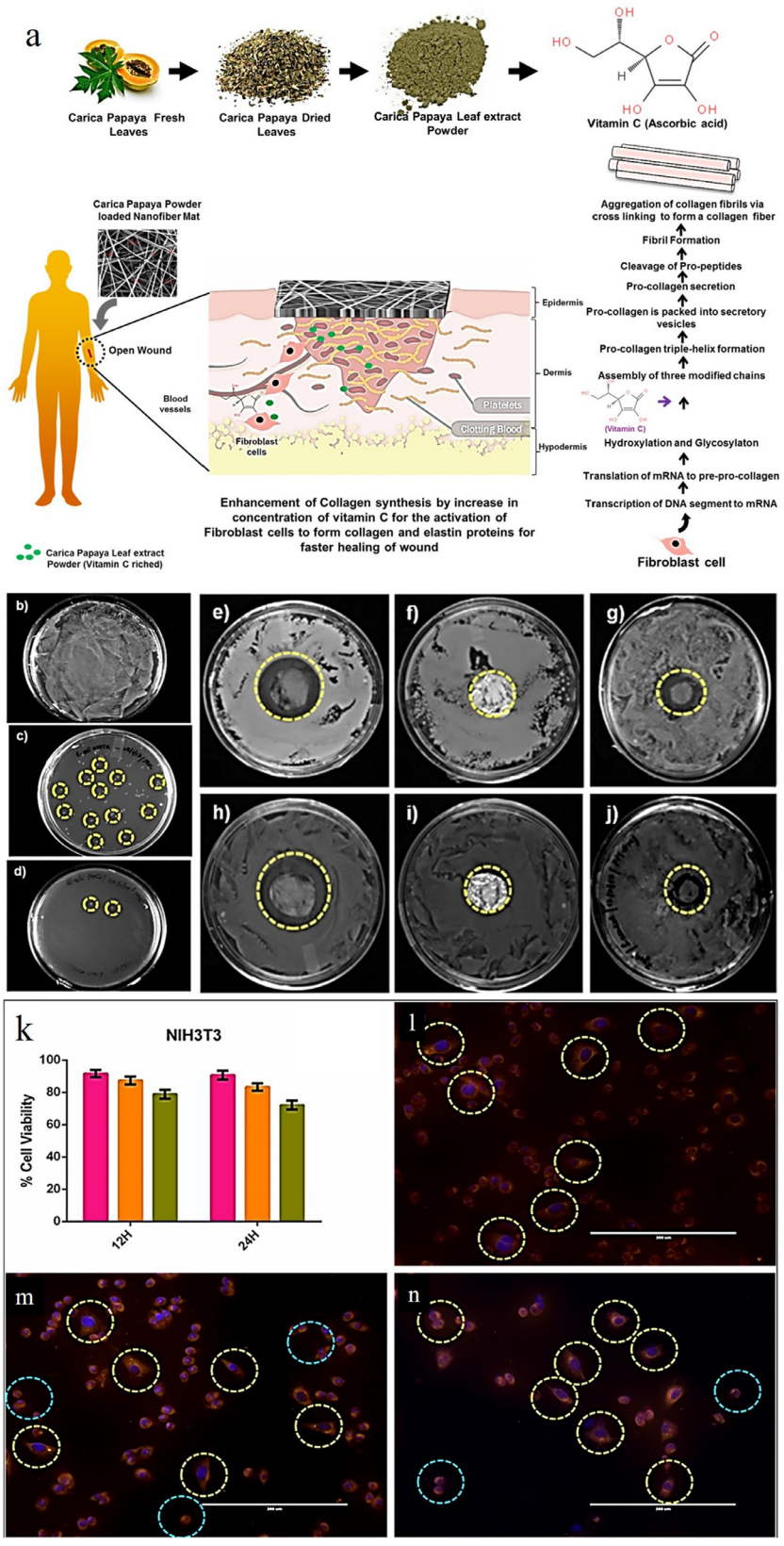

Fig. 10. a) Schematic illustration showing how Vitamin C present in *Carica papaya* leaf acts decisively in the process of wound healing. Antibacterial efficiency (CFU) of the *Carica papaya*/PVA/GE nanofibers (c and d: 10 mg and 12 mg of *Carica papaya*, respectively) versus that of the PVA/GE nanofibers (b) (the circles represent each bacterial colony). Antibacterial efficiency validated via disk diffusion assay for *Carica papaya* (e, h), PVA-GE nanofibers (f, i) and *Carica papaya* (12 mg)/PVA/GE nanofibers (g, j). The micrographs show the formed inhibition zone (marked by a circle) against *E. coli* (e–g) and *S. aureus* (h–j). (k) NIH 3T3 cell viability percentage when the cells are co-cultured with *Carica papaya*/ PVA/GE nanofibers. Two other columns represent the PVA/GE nanofibers as well as the nanofiber free medium. Cell morphology traced by Hoechst 33,342 and Rhodamine B staining of the cells co-cultured for 24 h with (l) nanofiber free medium; (m) PVA/GE nanofibers; and (n) *Carica papaya*/PVA/GE nanofibers (in the fluorescence microscopy images, the yellow and blue circles represent healthy cells and cytoplasmic constriction and nuclear fragmentation, respectively). Reproduced with permission [189]. Copyright 2019, Elsevier.

attention of the scientific community for the purpose of developing nanofibrous biomedical devices such as wound dressings. Given the large incidence of acute and chronic wounds across the world, the necessity of production of nanofibrous wound dressings able to control infection by releasing bactericidal agents and to block invasion of microbes is felt nowadays more than ever. Despite promising wound healing outcomes in terms of re-epithelialization, angiogenesis and antibacterial activity, nanofibrous wound dressings should be still improved in the following different aspects:

1- *Composition:* In terms of chemistry and composition, biopolymers that are biocompatible and induce anti-inflammatory and antibacterial properties, thereby accelerating wound healing, are preferred for nanofiber synthesis. Despite favorable potential of interactive biopolymeric nanofibrous dressings for wound healing, verified through many related studies, to the best of the authors' knowledge, there is no commercial product of them in the market. This arises from possible complications of electrospinning of biopolymers in a large scale, and biocompatibility concerns due to the existence of impurities such as cross-linkers and residual solvents in the fibers and the likely immunogenic reactions induced by such compounds. Specifically, biopolymers like chitosan and gelatine are hardly water soluble, thus for electrospinning they need to be dissolved in toxic, highly acidic solvents including 1,1,1,3,3,3-hexafluoro-2-propanol and trifluoroacetic acid (TFA) [190]. To acquire antimicrobial effect, the nanofibers should be equipped with secondary agents (additives) such as AMPs, metallic ions and nanoparticles, antibiotics, or plant-derived compounds, as discussed in this article. Taking into consideration the rapid evolution of antibiotic resistant bacteria, research is being carried out to develop alternatives to conventional drug delivery nanofibers, for example considering AMP functionalized ones. Recalling the hazardous dissolution and release of metallic (e.g. Ag) ions, if exceeding the WHO limits, both AMP functionalized nanofibers and plant-derived biohybrid nanofibers appear to be suitable substitutes for conventional drug delivery devices. However, long term *in vitro* and *in vivo* investigations of such systems are still demanded to guarantee their efficiency within the course of the wound healing process. To promote the wound healing effect, supplementary compounds such as growth factors, metallic ion delivering materials, etc. could be also embedded into the nanofibers. Furthermore, it is known that acidification of the wound milieu can accelerate the healing process, thus inclusion of various agents that help acidify the wound bed is another helpful strategy. Also, from the composition point of view, a state of the art class of smart nanofibrous wound dressings can be defined as those releasing antimicrobial agents and drugs when subjected to different stimuli including pH and temperature. This class of dressings has been rarely developed and studied and as prospective wound healing materials they should be further considered. Lastly, synthetic, industrial polymers could be also proposed for construction of wound dressings, provided that they show no toxicity effects. Such materials are promising due to their widely known processing methods, favorable physicochemical properties, the possibility of integration into engineered structures, and potential for scalability. These merits can be appealing for the development of wound dressing materials widely and economically. However, bioinertness is indeed a challenge that can complicate the removal process of the dressing upon healing of the wound. To address this bottleneck, a surface treatment involving biodegradable materials as a coating can be a solution.

2- *Synthesis:* With respect to antibacterial nanofibrous wound dressings, many antibiotics and anticancer drugs as well as antimicrobial agents have been conveniently incorporated into electrospun polymeric nanofibers for local delivery [42,117]. Traditionally, incorporation of such compounds into nanofibers is done via blending them into the polymer, followed by electrospinning of the blend or core-shell electrospinning wherein the drug/agent is located within a polymeric outer shell. In the former method, drugs or antimicrobial materials would loosely reside at the surface of the nanofibers, resulting in an unwanted burst release, thus imposing cytotoxicity toward tissue cells. In the latter one, involving high voltage and high shearing forces exerted at the interface between core and shell fluids, proteins could be rapidly dehydrated and delicate bioactive agents harmed. Thus, there is a need to develop alternative methods allowing for sustained delivery of antibacterial agents without damaging them. Despite versatility of electrospinning for production of nanofibers in different compositions and configurations, creation of an effective, reliable formulation for such biohybrid nanofibers is intricate. In this regard, the technique must be widely, yet precisely understood and standardized, particularly in the sophisticated versions of emulsion and coaxial electrospinning. Additionally, the production level of any desired formulation must be largely developed up to industrial scale. Hence, the production cycle must be economical and scalable, particularly considering the relatively low price of many available commercial wound dressings.

3- *Design and engineering:* Structurally, nanofibrous dressings need to be engineered so that either entrap bacteria or block their pathways into the wound. Moreover, coupled with nanoparticles, they can form a hierarchical nanostructure, whose nanotopography can potentially raise surface hydrophobicity thus lowering the chance of bacterial adhesion. As a result, biofilm formation and its adverse consequences can be drastically reduced. Moreover, nanofibrous dressings must hinder tissue ingrowth into the structure, enabling its easy and painless removal while encouraging cells to adhere and proliferate. Moreover, the dressing must be sufficiently porous and permeable to allow exchange of air and water vapor as well as nutrients and waste. On the other hand, such extent of porosity should not lead to loss of mechanical stability and pliability of the dressing. Given the diverse stresses applied to the dressing depending on its location and intended use duration, mechanical fatigue could be a crucial consideration as well. The dressing needs to adequately remove exudates, thus in addition to porosity, surface chemistry could play a vital role.

4- *Multifunctionality:* As a further necessity, the real-time monitoring of the wound bed conditions in terms of pH or temperature is a sophisticated challenge. Such parameters potentially indicate the wound healing status and they need to be assessed precisely and in the point of care. For this purpose, various classes of biosensors should be integrated into nanofibrous wound dressings and an advanced generation of smart devices, that are able to real time sense and monitor the wound conditions, should be developed. Accordingly, as an all in one package, multifunctional dressings are required that can treat various classes of chronic wounds while notably declining the infection tendency and wound recurrence. In fact, knowledge frontier should be pushed towards creation of dressings that merge three crucial functions, Fig. 11: (i) stimulating the healing process through the main, relevant physiological mechanisms and even by inclusion of further functionalities such as electrical responsiveness; (ii) monitoring the wound healing and infection indicators such as temperature, pH, and bacteria; and (iii) drug delivery in a controlled manner if wound infection emerges. A nanofibrous wound dressing fulfilling these concerns guarantees not only fast healing of a wound but also avoids infection occurrence. Moreover, the treatment cycle and duration could be predicted and controlled over time by

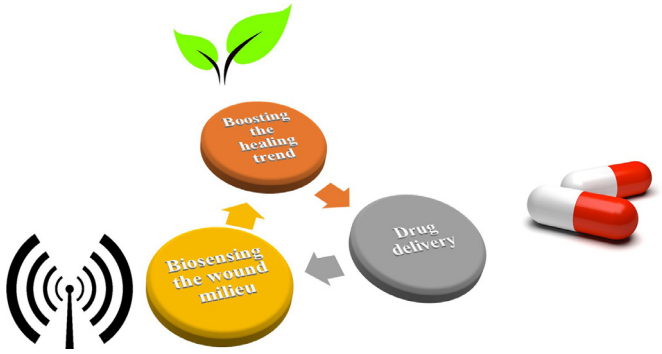

Fig. 11. Three main requirements for the next generation of nanofibrous wound dressings (the images have been obtained from http://www.Bing.com and are under Creative Commons Licence)

providing diverse readouts by the integrated biosensor concerning wound healing status, exudate amount, infection, and lifespan of the dressing [5].

5- *Testing:* Alongside production, advanced testing approaches for the developed nanofibrous systems must be designed and verified to enable reliable evaluation and prompt translation of these devices to clinical applications. For instance, to determine the clinical potential of such dressings, the *in vitro* interaction of the nanofibers with cells must be investigated in the presence of bacteria to simulate realistically the conditions of the wound beds. Surprisingly, almost no research in the literature has considered this important characterization involving bacteria and cell co-cultures. The achieved information can shed light on the adhesion and growth mechanisms of bacteria onto the nanofibers, thereby enabling physically or chemically adjustment of their characteristics to overcome microbial challenges. Consequently, the impact of nanofibrous dressings on dermal cells in the presence of bacteria should be largely investigated to assure applicability of the dressings in microbiological environments similar to those of real wounds. Certainly, depending on the type of wound, involving various specific biological factors, and its chronicity, *in vitro* tests need to be customized. In addition to the development of advanced biological testing, physicochemical characteristics must be investigated under realistic conditions in terms of pH, humidity, temperature, mechanical and thermal stress magnitude and frequency to simulate the practical situation. In this case, it is necessary to follow the designed standards for such kind of materials to enable comparison with the available benchmarks and to verify their modifications.

Declaration of Competing Interest

The authors declare no conflict of interest.

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References

[1] http://cordis.europa.eu/result/rcn/92892_en.html.
[2] C.K. Sen, G.M. Gordillo, S. Roy, R. Kirsner, L. Lambert, T.K. Hunt, F. Gottrup, G.C. Gurtner, M.T. Longaker, Human skin wounds: a major and snowballing threat to public health and the economy, Wound Repair Regen. 17 (6) (2009) 763–771.
[3] H.P. Felgueiras, M.T.P. Amorim, Functionalization of electrospun polymeric wound dressings with antimicrobial peptides, Colloids Surf. B: Biointerfaces 156 (2017) 133–148.
[4] http://www.marketsandmarkets.com/Market-Reports/wound-care-market-371.html.
[5] M. Abrigo, S.L. McArthur, P. Kingshott, Electrospun nanofibers as dressings for chronic wound care: advances, challenges, and future prospects, Macromol. Biosci. 14 (6) (2014) 772–792.
[6] T. Ondarcuhu, C. Joachim, Drawing a single nanofibre over hundreds of microns, Europhys. Lett. 42 (2) (1998) 215.
[7] L. Feng, S. Li, H. Li, J. Zhai, Y. Song, L. Jiang, D. Zhu, Super-hydrophobic surface of aligned polyacrylonitrile nanofibers, Angew. Chem. Int. Ed. 41 (7) (2002) 1221–1223.
[8] P.X. Ma, R. Zhang, Synthetic nano-scale fibrous extracellular matrix, J. Biomed. Mater. Res. 46 (1) (1999) 60–72.
[9] G.M. Whitesides, B. Grzybowski, Self-assembly at all scales, Science 295 (5564) (2002) 2418–2421.
[10] S. Homaeigohar, T. Dai, M. Elbahri, Biofunctionalized nanofibrous membranes as super separators of protein and enzyme from water, J. Colloid Interface Sci. 406 (2013) 86–93.
[11] S. Homaeigohar, D. Disci-Zayed, T. Dai, M. Elbahri, Biofunctionalized nanofibrous membranes mimicking carnivorous plants, Bioinspir., Biomim. Nanobiomater. 2 (4) (2013) 186–193.
[12] S. Homaeigohar, A.U. Zillohu, R. Abdelaziz, M.K. Hedayati, M. Elbahri, A novel nanohybrid nanofibrous adsorbent for water purification from dye pollutants, Materials 9 (10) (2016) 848.
[13] M. Elbahri, S. Homaeigohar, T. Dai, R. Abdelaziz, R. Khalil, A.U. Zillohu, Smart metal–polymer bionanocomposites as omnidirectional plasmonic black absorbers formed by nanofluid filtration, Adv. Funct. Mater. 22 (22) (2012) 4771–4777.
[14] S.S. Homaeigohar, M. Elbahri, Novel compaction resistant and ductile nanocomposite nanofibrous microfiltration membranes, J. Colloid Interface Sci. 372 (1) (2012) 6–15.
[15] S.S. Homaeigohar, H. Mahdavi, M. Elbahri, Extraordinarily water permeable sol–gel formed nanocomposite nanofibrous membranes, J. Colloid Interface Sci. 366 (2012) 51–56.
[16] S. Homaeigohar, M. Elbahri, Nanocomposite electrospun nanofiber membranes for environmental remediation, Materials 7 (2) (2014) 1017–1045.
[17] Z.-M. Huang, Y.Z. Zhang, M. Kotaki, S. Ramakrishna, A review on polymer nanofibers by electrospinning and their applications in nanocomposites, Compos. Sci. Technol. 63 (15) (2003) 2223–2253.
[18] V. Andreu, G. Mendoza, M. Arruebo, S. Irusta, Smart dressings based on nanostructured fibers containing natural origin antimicrobial, anti-inflammatory, and regenerative compounds, Materials 8 (8) (2015) 5154–5193.
[19] P. Zahedi, I. Rezaeian, S.O. Ranaei-Siadat, S.H. Jafari, P. Supaphol, A review on wound dressings with an emphasis on electrospun nanofibrous polymeric bandages, Polym. Adv. Technol. 21 (2) (2010) 77–95.
[20] M. Liu, X.-P. Duan, Y.-M. Li, D.-P. Yang, Y.-Z. Long, Electrospun nanofibers for wound healing, Mater. Sci. Eng.: C 76 (2017) 1413–1423.
[21] D. Simões, S.P. Miguel, M.P. Ribeiro, P. Coutinho, A.G. Mendonça, I.J. Correia, Recent advances on antimicrobial wound dressing: a review, Eur. J. Pharm. Biopharm. 127 (2018) 130–141.
[22] J. Wang, M. Windbergs, Functional electrospun fibers for the treatment of human skin wounds, Eur. J. Pharm. Biopharm. 119 (2017) 283–299.
[23] Y. Liu, S. Zhou, Y. Gao, Y. Zhai, Electrospun nanofibers as a wound dressing for treating diabetic foot ulcer, Asian J. Pharm. Sci. 14 (2) (2019) 130–143.
[24] S.P. Miguel, D.R. Figueira, D. Simões, M.P. Ribeiro, P. Coutinho, P. Ferreira, I.J. Correia, Electrospun polymeric nanofibres as wound dressings: a review, Colloids Surf. B: Biointerfaces 169 (2018) 60–71.
[25] S. Chen, B. Liu, M.A. Carlson, A.F. Gombart, D.A. Reilly, J. Xie, Recent advances in electrospun nanofibers for wound healing, Nanomedicine 12 (11) (2017) 1335–1352.
[26] S. Torres-Giner, R. Pérez-Masiá, J.M. Lagaron, A review on electrospun polymer nanostructures as advanced bioactive platforms, Polym. Eng. Sci. 56 (5) (2016) 500–527.
[27] R.S. Ambekar, B. Kandasubramanian, Advancements in nanofibers for wound dressing: a review, Eur. Polym. J. 117 (2019) 304–336.
[28] S. Fahimirad, F. Ajalloueian, Naturally-derived electrospun wound dressings for target delivery of bio-active agents, Int. J. Pharm. 566 (2019) 307–328.
[29] M. Naseri-Nosar, Z.M. Ziora, Wound dressings from naturally-occurring polymers: a review on homopolysaccharide-based composites, Carbohydr. Polym. 189 (2018) 379–398.
[30] S.P. Miguel, R.S. Sequeira, A.F. Moreira, C.S.D. Cabral, A.G. Mendonça, P. Ferreira, I.J. Correia, An overview of electrospun membranes loaded with bioactive molecules for improving the wound healing process, Eur. J. Pharm. Biopharm. 139 (2019) 1–22.
[31] H. Rodríguez-Tobías, G. Morales, D. Grande, Comprehensive review on electrospinning techniques as versatile approaches toward antimicrobial biopolymeric composite fibers, Mater. Sci. Eng.: C 101 (2019) 306–322.
[32] G. Han, R. Ceilley, Chronic wound healing: a review of current management and treatments, Adv. Ther. 34 (3) (2017) 599–610.
[33] T.A. Wynn, L. Barron, in: Macrophages: Master Regulators of Inflammation and Fibrosis, Seminars in Liver Disease, Thieme Medical Publishers, 2010, pp. 245–257.
[34] P. Martin, R. Nunan, Cellular and molecular mechanisms of repair in acute and chronic wound healing, Brit. J. Dermatol. 173 (2) (2015) 370–378.

[35] B. Eckes, R. Nischt, T. Krieg, Cell-matrix interactions in dermal repair and scarring, Fibrogenesis Tissue Repair 3 (1) (2010) 4.
[36] F.G. Giancotti, E. Ruoslahti, Integrin signaling, Science 285 (5430) (1999) 1028–1033.
[37] L. Koivisto, J. Heino, L. Häkkinen, H. Larjava, Integrins in wound healing, Adv. Wound Care 3 (12) (2014) 762–783.
[38] S. Barrientos, O. Stojadinovic, M.S. Golinko, H. Brem, M. Tomic-Canic, Growth factors and cytokines in wound healing, Wound Repair Regen. 16 (5) (2008) 585–601.
[39] K. Hyldig, S. Riis, C.P. Pennisi, V. Zachar, T. Fink, Implications of extracellular matrix production by adipose tissue-derived stem cells for development of wound healing therapies, Int. J. Mol. Sci. 18 (6) (2017) 1167.
[40] K. Haukipuro, J. Melkko, L. Risteli, M. Kairaluoma, J. Risteli, Synthesis of type I collagen in healing wounds in humans, Ann. Surg. 213 (1) (1991) 75.
[41] G.C. Gurtner, S. Werner, Y. Barrandon, M.T. Longaker, Wound repair and regeneration, Nature 453 (7193) (2008) 314.
[42] A.C. Alavarse, F.W. de Oliveira Silva, J.T. Colque, V.M. da Silva, T. Prieto, E.C. Venancio, J.-J. Bonvent, Tetracycline hydrochloride-loaded electrospun nanofibers mats based on PVA and chitosan for wound dressing, Mater. Sci. Eng.: C 77 (2017) 271–281.
[43] D. Chouhan, B. Chakraborty, S.K. Nandi, B.B. Mandal, Role of non-mulberry silk fibroin in deposition and regulation of extracellular matrix towards accelerated wound healing, Acta Biomater. 48 (2017) 157–174.
[44] T. Bjarnsholt, K. Kirketerp-Møller, P.Ø. Jensen, K.G. Madsen, R. Phipps, K. Krogfelt, N. Høiby, M. Givskov, Why chronic wounds will not heal: a novel hypothesis, Wound Repair Regen. 16 (1) (2008) 2–10.
[45] K.A. Rieger, N.P. Birch, J.D. Schiffman, Designing electrospun nanofiber mats to promote wound healing—a review, J. Mater. Chem. B 1 (36) (2013) 4531–4541.
[46] J.M. Smiell, T.J. Wieman, D.L. Steed, B.H. Perry, A.R. Sampson, B.H. Schwab, Efficacy and safety of becaplermin (recombinant human platelet-derived growth factor-BB) in patients with nonhealing, lower extremity diabetic ulcers: a combined analysis of four randomized studies, Wound Repair Regen. 7 (5) (1999) 335–346.
[47] B.A. Lipsky, C. Hoey, Topical antimicrobial therapy for treating chronic wounds, Clin. Infect. Dis. 49 (10) (2009) 1541–1549.
[48] J.G. Powers, C. Higham, K. Broussard, T.J. Phillips, Wound healing and treating wounds: chronic wound care and management, J. Am. Acad. Dermatol. 74 (4) (2016) 607–625.
[49] W.A. Marston, J. Hanft, P. Norwood, R. Pollak, The efficacy and safety of Dermagraft in improving the healing of chronic diabetic foot ulcers: results of a prospective randomized trial, Diabetes Care 26 (6) (2003) 1701–1705.
[50] L.I. Moura, A.M. Dias, E. Carvalho, H.C. de Sousa, Recent advances on the development of wound dressings for diabetic foot ulcer treatment—a review, Acta Biomater. 9 (7) (2013) 7093–7114.
[51] S.Z. Fu, X.H. Meng, J. Fan, L.L. Yang, Q.L. Wen, S.J. Ye, S. Lin, B.Q. Wang, L.L. Chen, J.B. Wu, Acceleration of dermal wound healing by using electrospun curcumin-loaded poly (ε-caprolactone)-poly (ethylene glycol)-poly (ε-caprolactone) fibrous mats, J. Biomed. Mater. Res. B: Appl. Biomater. 102 (3) (2014) 533–542.
[52] L.G. Ovington, Advances in wound dressings, Clin. Dermatol. 25 (1) (2007) 33–38.
[53] G.T. Lionelli, W.T. Lawrence, Wound dressings, Surg. Clin. 83 (3) (2003) 617–638.
[54] E. Fiume, J. Barberi, E. Verné, F. Baino, Bioactive glasses: from parent 45S5 composition to scaffold-assisted tissue-healing therapies, J. Funct. Biomater. 9 (1) (2018) 24.
[55] T. Coats, C. Edwards, R. Newton, E. Staun, The effect of gel burns dressings on skin temperature, Emerg. Med. J. 19 (3) (2002) 224–225.
[56] S. Huang, X. Fu, Naturally derived materials-based cell and drug delivery systems in skin regeneration, J. Control. Rel. 142 (2) (2010) 149–159.
[57] G.D. Mogoşanu, A.M. Grumezescu, Natural and synthetic polymers for wounds and burns dressing, Int. J. Pharm. 463 (2) (2014) 127–136.
[58] T. Zhou, N. Wang, Y. Xue, T. Ding, X. Liu, X. Mo, J. Sun, Electrospun tilapia collagen nanofibers accelerating wound healing via inducing keratinocytes proliferation and differentiation, Colloids Surf. B: Biointerfaces 143 (2016) 415–422.
[59] W. Czaja, A. Krystynowicz, S. Bielecki, R.M. Brown Jr., Microbial cellulose—the natural power to heal wounds, Biomaterials 27 (2) (2006) 145–151.
[60] T. Hakkarainen, R. Koivuniemi, M. Kosonen, C. Escobedo-Lucea, A. Sanz-Garcia, J. Vuola, J. Valtonen, P. Tammela, A. Mäkitie, K. Luukko, M. Yliperttula, H. Kavola, Nanofibrillar cellulose wound dressing in skin graft donor site treatment, J. Control. Release 244 (2016) 292–301.
[61] W.K. Czaja, D.J. Young, M. Kawecki, R.M. Brown, The future prospects of microbial cellulose in biomedical applications, Biomacromolecules 8 (1) (2007) 1–12.
[62] P. Wutticharoenmongkol, P. Hannirojram, P. Nuthong, Gallic acid-loaded electrospun cellulose acetate nanofibers as potential wound dressing materials, Polym. Adv. Technol. 30 (4) (2019) 1135–1147.
[63] Y. Liu, G. Ma, D. Fang, J. Xu, H. Zhang, J. Nie, Effects of solution properties and electric field on the electrospinning of hyaluronic acid, Carbohydr. Polym. 83 (2) (2011) 1011–1015.
[64] J. Baier Leach, K.A. Bivens, C.W. Patrick Jr., C.E. Schmidt, Photocrosslinked hyaluronic acid hydrogels: natural, biodegradable tissue engineering scaffolds, Biotechnol. Bioeng. 82 (5) (2003) 578–589.
[65] D. Eng, M. Caplan, M. Preul, A. Panitch, Hyaluronan scaffolds: a balance between backbone functionalization and bioactivity, Acta Biomater. 6 (7) (2010) 2407–2414.
[66] W. Chen, Preparation of hyaluronan-DNA matrices and films, Cold Spring Harb. Protoc. 2012 (10) (2012) pdb. prot071522.
[67] R. Uppal, G.N. Ramaswamy, C. Arnold, R. Goodband, Y. Wang, Hyaluronic acid nanofiber wound dressing—production, characterization, and *in vivo* behavior, J. Biomed. Mater. Res. B: Appl. Biomater. 97 (1) (2011) 20–29.
[68] Y.C. Shin, D.-M. Shin, E.J. Lee, J.H. Lee, J.E. Kim, S.H. Song, D.-Y. Hwang, J.J. Lee, B. Kim, D. Lim, S.-H. Hyon, Y.-J. Lim, D.-W. Han, Hyaluronic acid/PLGA core/shell fiber matrices loaded with EGCG beneficial to diabetic wound healing, Adv. Healthc. Mater. 5 (23) (2016) 3035–3045.
[69] M. Séon-Lutz, A.-C. Couffin, S. Vignoud, G. Schlatter, A. Hébraud, Electrospinning in water and in situ crosslinking of hyaluronic acid / cyclodextrin nanofibers: towards wound dressing with controlled drug release, Carbohydr. Polym. 207 (2019) 276–287.
[70] N. Bhardwaj, S.C. Kundu, Electrospinning: a fascinating fiber fabrication technique, Biotechnol. Adv. 28 (3) (2010) 325–347.
[71] S.-J. Liu, Y.-C. Kau, C.-Y. Chou, J.-K. Chen, R.-C. Wu, W.-L. Yeh, Electrospun PLGA/collagen nanofibrous membrane as early-stage wound dressing, J. Memb. Sci. 355 (1–2) (2010) 53–59.
[72] J. Gunn, M. Zhang, Polyblend nanofibers for biomedical applications: perspectives and challenges, Trends Biotechnol. 28 (4) (2010) 189–197.
[73] G. Panthi, N.A. Barakat, P. Risal, A. Yousef, B. Pant, A.R. Unnithan, H.Y. Kim, Preparation and characterization of nylon-6/gelatin composite nanofibers via electrospinning for biomedical applications, Fibers Polym. 14 (5) (2013) 718–723.
[74] A.M. Abdelgawad, S.M. Hudson, O.J. Rojas, Antimicrobial wound dressing nanofiber mats from multicomponent (chitosan/silver-NPs/polyvinyl alcohol) systems, Carbohydr. Polym. 100 (2014) 166–178.
[75] A. Mohandas, B. Anisha, K. Chennazhi, R. Jayakumar, Chitosan–hyaluronic acid/VEGF loaded fibrin nanoparticles composite sponges for enhancing angiogenesis in wounds, Colloids Surf. B: Biointerfaces 127 (2015) 105–113.
[76] W. Paul, C.P. Sharma, Chitosan and alginate wound dressings: a short review, Trends Biomater. Artif. Organs 18 (1) (2004) 18–23.
[77] M. Dash, F. Chiellini, R.M. Ottenbrite, E. Chiellini, Chitosan—A versatile semi-synthetic polymer in biomedical applications, Prog. Polym. Sci. 36 (8) (2011) 981–1014.
[78] H. Adeli, M.T. Khorasani, M. Parvazinia, Wound dressing based on electrospun PVA/chitosan/starch nanofibrous mats: fabrication, antibacterial and cytocompatibility evaluation and *in vitro* healing assay, Int. J. Biol. Macromol. 122 (2019) 238–254.
[79] R. Ahmed, M. Tariq, I. Ali, R. Asghar, P. Noorunnisa Khanam, R. Augustine, A. Hasan, Novel electrospun chitosan/polyvinyl alcohol/zinc oxide nanofibrous mats with antibacterial and antioxidant properties for diabetic wound healing, Int. J. Biol. Macromol. 120 (2018) 385–393.
[80] B.P. Antunes, A.F. Moreira, V.M. Gaspar, I.J. Correia, Chitosan/arginine–chitosan polymer blends for assembly of nanofibrous membranes for wound regeneration, Carbohydr. Polym. 130 (2015) 104–112.
[81] W.A. Sarhan, H.M.E. Azzazy, I.M. El-Sherbiny, Honey/chitosan nanofiber wound dressing enriched with allium sativum and cleome droserifolia: enhanced antimicrobial and wound healing activity, ACS Appl. Mater. Interfaces 8 (10) (2016) 6379–6390.
[82] N. Ardila, N. Medina, M. Arkoun, M.-C. Heuzey, A. Ajji, C.J. Panchal, Chitosan–bacterial nanocellulose nanofibrous structures for potential wound dressing applications, Cellulose 23 (5) (2016) 3089–3104.
[83] K. Kalantari, A.M. Afifi, H. Jahangirian, T.J. Webster, Biomedical applications of chitosan electrospun nanofibers as a green polymer – review, Carbohydr. Polym. 207 (2019) 588–600.
[84] S. Homaeigohar, T.-Y. Tsai, T.-H. Young, H.J. Yang, Y.-R. Ji, An electroactive alginate hydrogel nanocomposite reinforced by functionalized graphite nanofilaments for neural tissue engineering, Carbohydr. Polym. (2019) 115112.
[85] K.Y. Lee, D.J. Mooney, Alginate: properties and biomedical applications, Prog. Polym. Sci. 37 (1) (2012) 106–126.
[86] C.H. Goh, P.W.S. Heng, L.W. Chan, Alginates as a useful natural polymer for microencapsulation and therapeutic applications, Carbohydr. Polym. 88 (1) (2012) 1–12.
[87] Y. Tang, X. Lan, C. Liang, Z. Zhong, R. Xie, Y. Zhou, X. Miao, H. Wang, W. Wang, Honey loaded alginate/PVA nanofibrous membrane as potential bioactive wound dressing, Carbohydr. Polym. 219 (2019) 113–120.
[88] H.P. Felgueiras, N.S. Murthy, S.D. Sommerfeld, M.M. Brás, V. Migonney, J. Kohn, Competitive adsorption of plasma proteins using a quartz crystal microbalance, ACS Appl. Mater. Interfaces 8 (21) (2016) 13207–13217.
[89] M. Albu, M. Ferdes, D. Kaya, M. Ghica, I. Titorencu, L. Popa, L. Albu, Collagen wound dressings with anti-inflammatory activity, Mol. Cryst. Liq. Cryst. 555 (1) (2012) 271–279.
[90] D. Brett, A review of collagen and collagen-based wound dressings, Wounds 20 (12) (2008) 347–356.
[91] X. Wen, Y. Zheng, J. Wu, L.-N. Wang, Z. Yuan, J. Peng, H. Meng, Immobilization of collagen peptide on dialdehyde bacterial cellulose nanofibers via covalent bonds for tissue engineering and regeneration, Int. J. Nanomed. 10 (2015) 4623.
[92] A. Karthika, L. Kavitha, M. Surendiran, S. Kannan, D. Gopi, Fabrication of divalent ion substituted hydroxyapatite/gelatin nanocomposite coating on electron beam treated titanium: mechanical, anticorrosive, antibacterial and bioactive evaluations, RSC Adv. 5 (59) (2015) 47341–47352.
[93] C.-H. Yao, C.-Y. Lee, C.-H. Huang, Y.-S. Chen, K.-Y. Chen, Novel bilayer wound dressing based on electrospun gelatin/keratin nanofibrous mats for skin wound repair, Mater. Sci. Eng.: C 79 (2017) 533–540.

[94] J. Melke, S. Midha, S. Ghosh, K. Ito, S. Hofmann, Silk fibroin as biomaterial for bone tissue engineering, Acta Biomater. 31 (2016) 1–16.
[95] A. Gupta, K. Mita, K.P. Arunkumar, J. Nagaraju, Molecular architecture of silk fibroin of Indian golden silkmoth, *Antheraea assama*, Sci. Rep. 5 (2015) 12706.
[96] L. Fan, Z. Cai, K. Zhang, F. Han, J. Li, C. He, X. Mo, X. Wang, H. Wang, Green electrospun pantothenic acid/silk fibroin composite nanofibers: fabrication, characterization and biological activity, Colloids Surf. B: Biointerfaces 117 (2014) 14–20.
[97] D.W. Song, S.H. Kim, H.H. Kim, K.H. Lee, C.S. Ki, Y.H. Park, Multi-biofunction of antimicrobial peptide-immobilized silk fibroin nanofiber membrane: implications for wound healing, Acta Biomater. 39 (2016) 146–155.
[98] X. Yang, L. Fan, L. Ma, Y. Wang, S. Lin, F. Yu, X. Pan, G. Luo, D. Zhang, H. Wang, Green electrospun Manuka honey/silk fibroin fibrous matrices as potential wound dressing, Mater. Des. 119 (2017) 76–84.
[99] S. Selvaraj, N.N. Fathima, Fenugreek incorporated silk fibroin nanofibers—a potential antioxidant scaffold for enhanced wound healing, ACS Appl. Mater. Interfaces 9 (7) (2017) 5916–5926.
[100] M. Farokhi, F. Mottaghitalab, Y. Fatahi, A. Khademhosseini, D.L. Kaplan, Overview of silk fibroin use in wound dressings, Trends Biotechnol. 36 (9) (2018) 907–922.
[101] L.-D. Koh, J. Yeo, Y.Y. Lee, Q. Ong, M. Han, B.C.K. Tee, Advancing the frontiers of silk fibroin protein-based materials for futuristic electronics and clinical wound-healing (Invited review), Mater. Sci. Eng.: C 86 (2018) 151–172.
[102] R. Edwards, K.G. Harding, Bacteria and wound healing, Curr. Opin. Infect. Dis. 17 (2) (2004) 91–96.
[103] A.R. Siddiqui, J.M. Bernstein, Chronic wound infection: facts and controversies, Clin. Dermatol. 28 (5) (2010) 519–526.
[104] F. Siedenbiedel, J.C. Tiller, Antimicrobial polymers in solution and on surfaces: overview and functional principles, Polymers 4 (1) (2012) 46–71.
[105] M.V. Park, A.M. Neigh, J.P. Vermeulen, L.J. de la Fonteyne, H.W. Verharen, J.J. Briedé, H. van Loveren, W.H. de Jong, The effect of particle size on the cytotoxicity, inflammation, developmental toxicity and genotoxicity of silver nanoparticles, Biomaterials 32 (36) (2011) 9810–9817.
[106] R.E. Hancock, H.-G. Sahl, Antimicrobial and host-defense peptides as new anti-infective therapeutic strategies, Nat. Biotechnol. 24 (12) (2006) 1551.
[107] M. Zasloff, Antimicrobial peptides of multicellular organisms, Nature 415 (6870) (2002) 389.
[108] A. Parisien, B. Allain, J. Zhang, R. Mandeville, C. Lan, Novel alternatives to antibiotics: bacteriophages, bacterial cell wall hydrolases, and antimicrobial peptides, J. Appl. Microbiol. 104 (1) (2008) 1–13.
[109] R. Ramos, J.P. Silva, A.C. Rodrigues, R. Costa, L. Guardão, F. Schmitt, R. Soares, M. Vilanova, L. Domingues, M. Gama, Wound healing activity of the human antimicrobial peptide LL37, Peptides 32 (7) (2011) 1469–1476.
[110] T. Akiyama, F. Niyonsaba, C. Kiatsurayanon, H. Ushio, T. Fujimura, T. Ueno, K. Okumura, H. Ogawa, S. Ikeda, The human cathelicidin LL-37 host defense peptide upregulates tight junction-related proteins and increases human epidermal keratinocyte barrier function, J. Innate Immun. 6 (6) (2014) 739–753.
[111] A.K.T. Chang, R.R. Frias, L.V. Alvarez, U.G. Bigol, J.P.M.D. Guzman, Comparative antibacterial activity of commercial chitosan and chitosan extracted from Auricularia sp, Biocatal. Agric. Biotechnol. 17 (2019) 189–195.
[112] M. Jannesari, J. Varshosaz, M. Morshed, M. Zamani, Composite poly (vinyl alcohol)/poly (vinyl acetate) electrospun nanofibrous mats as a novel wound dressing matrix for controlled release of drugs, Int. J. Nanomed. 6 (2011) 993.
[113] X. Hu, S. Liu, G. Zhou, Y. Huang, Z. Xie, X. Jing, Electrospinning of polymeric nanofibers for drug delivery applications, J. Control. Release 185 (2014) 12–21.
[114] W. Cui, Y. Zhou, J. Chang, Electrospun nanofibrous materials for tissue engineering and drug delivery, Sci. Technol. Adv. Mater. 11 (1) (2010) 014108.
[115] H. Jiang, L. Wang, K. Zhu, Coaxial electrospinning for encapsulation and controlled release of fragile water-soluble bioactive agents, J. Control. Release 193 (0) (2014) 296–303.
[116] N. Monteiro, M. Martins, A. Martins, N.A. Fonseca, J.N. Moreira, R.L. Reis, N.M. Neves, Antibacterial activity of chitosan nanofiber meshes with liposomes immobilized releasing gentamicin, Acta Biomater. 18 (2015) 196–205.
[117] G. Rath, T. Hussain, G. Chauhan, T. Garg, A.K. Goyal, Development and characterization of cefazolin loaded zinc oxide nanoparticles composite gelatin nanofiber mats for postoperative surgical wounds, Mater. Sci. Eng.: C 58 (2016) 242–253.
[118] N. Liao, A.R. Unnithan, M.K. Joshi, A.P. Tiwari, S.T. Hong, C.-H. Park, C.S. Kim, Electrospun bioactive poly (ε-caprolactone)–cellulose acetate–dextran antibacterial composite mats for wound dressing applications, Colloids Surf. A: Physicochem. Eng. Asp. 469 (2015) 194–201.
[119] S. Chao, Y. Li, R. Zhao, L. Zhang, Y. Li, C. Wang, X. Li, Synthesis and characterization of tigecycline-loaded sericin/poly(vinyl alcohol) composite fibers via electrospinning as antibacterial wound dressings, J. Drug Deliv. Sci. Technol. 44 (2018) 440–447.
[120] F.A. Paskiabi, S. Bonakdar, M.A. Shokrgozar, M. Imani, Z. Jahanshiri, M. Shams-Ghahfarokhi, M. Razzaghi-Abyaneh, Terbinafine-loaded wound dressing for chronic superficial fungal infections, Mater. Sci. Eng.: C 73 (2017) 130–136.
[121] H. Li, Z. Zhang, V.U. Godakanda, Y.-J. Chiu, U. Angkawinitwong, K. Patel, P.G. Stapleton, R.M. de Silva, K.M.N. de Silva, L.-M. Zhu, G.R. Williams, The effect of collection substrate on electrospun ciprofloxacin-loaded poly(vinylpyrrolidone) and ethyl cellulose nanofibers as potential wound dressing materials, Mater. Sci. Eng.: C 104 (2019) 109917.
[122] D. Semnani, N. Poursharifi, N. Banitaba, A. Fakhrali, Electrospun polyvinylidene pyrolidone/gelatin membrane impregnated with silver sulfadiazine as wound dressing for burn treatment, Bull. Mater. Sci. 41 (3) (2018) 72.
[123] M. Safdari, E. Shakiba, S.H. Kiaie, A. Fattahi, Preparation and characterization of Ceftazidime loaded electrospun silk fibroin/gelatin mat for wound dressing, Fibers Polym. 17 (5) (2016) 744–750.
[124] T. Xia, M. Kovochich, M. Liong, L. Mädler, B. Gilbert, H. Shi, J.I. Yeh, J.I. Zink, A.E. Nel, Comparison of the mechanism of toxicity of zinc oxide and cerium oxide nanoparticles based on dissolution and oxidative stress properties, ACS Nano 2 (10) (2008) 2121–2134.
[125] R. Fu, C. Li, C. Yu, H. Xie, S. Shi, Z. Li, Q. Wang, L. Lu, A novel electrospun membrane based on moxifloxacin hydrochloride/poly(vinyl alcohol)/sodium alginate for antibacterial wound dressings in practical application, Drug Deliv. 23 (3) (2016) 818–829.
[126] C. Ma, L. Liu, W. Hua, Y. Cai, J. Yao, Fabrication and characterization of absorbent and antibacterial alginate fibers loaded with sulfanilamide, Fibers Polym. 16 (6) (2015) 1255–1261.
[127] M.M. Stevens, J.H. George, Exploring and engineering the cell surface interface, Science 310 (5751) (2005) 1135–1138.
[128] M.A. Fonder, G.S. Lazarus, D.A. Cowan, B. Aronson-Cook, A.R. Kohli, A.J. Mamelak, Treating the chronic wound: a practical approach to the care of nonhealing wounds and wound care dressings, J. Am. Acad. Dermatol. 58 (2) (2008) 185–206.
[129] F.-L. Mi, S.-S. Shyu, Y.-B. Wu, S.-T. Lee, J.-Y. Shyong, R.-N. Huang, Fabrication and characterization of a sponge-like asymmetric chitosan membrane as a wound dressing, Biomaterials 22 (2) (2001) 165–173.
[130] A. GhavamiNejad, A. Rajan Unnithan, A. Ramachandra Kurup Sasikala, M. Samarikhalaj, R.G. Thomas, Y.Y. Jeong, S. Nasseri, P. Murugesan, D. Wu, C. Hee Park, C.S. Kim, Mussel-Inspired electrospun nanofibers functionalized with size-controlled silver nanoparticles for wound dressing application, ACS Appl. Mater. Interfaces 7 (22) (2015) 12176–12183.
[131] B.S. Atiyeh, M. Costagliola, S.N. Hayek, S.A. Dibo, Effect of silver on burn wound infection control and healing: review of the literature, Burns 33 (2) (2007) 139–148.
[132] A. Russell, W. Hugo, 7 Antimicrobial activity and action of silver, Progr. Med. Chem. 31 (1994) 351–370.
[133] F.-R.F. Fan, A.J. Bard, Chemical, electrochemical, gravimetric, and microscopic studies on antimicrobial silver films, J. Phys. Chem. B 106 (2) (2002) 279–287.
[134] J.B. Wright, K. Lam, R.E. Burrell, Wound management in an era of increasing bacterial antibiotic resistance: a role for topical silver treatment, Am. J. Infect. Control 26 (6) (1998) 572–577.
[135] A.B. Lansdown, The role of silver, European Tissue Repair Society Bulletin (2002).
[136] S. Homaeigohar, N.K. Botcha, E.S. Zarie, M. Elbahri, Ups and downs of water photodecolorization by nanocomposite polymer nanofibers, Nanomaterials 9 (2) (2019) 250.
[137] S. Homaeigohar, Y. Davoudpour, Y. Habibi, M. Elbahri, The electrospun ceramic hollow nanofibers, Nanomaterials 7 (11) (2017) 383.
[138] H. Lee, S.M. Dellatore, W.M. Miller, P.B. Messersmith, Mussel-Inspired surface chemistry for multifunctional coatings, Science 318 (5849) (2007) 426–430.
[139] J.-L. Wang, B.-C. Li, Z.-J. Li, K.-F. Ren, L.-J. Jin, S.-M. Zhang, H. Chang, Y.-X. Sun, J. Ji, Electropolymerization of dopamine for surface modification of complex-shaped cardiovascular stents, Biomaterials 35 (27) (2014) 7679–7689.
[140] C.S. Costa, J.V.V. Ronconi, J.F. Daufenbach, C.L. Gonçalves, G.T. Rezin, E.L. Streck, M.M. da Silva Paula, *in vitro* effects of silver nanoparticles on the mitochondrial respiratory chain, Mol. Cell. Biochem. 342 (1/2) (2010) 51–56.
[141] R. Nudelman, H. Alhmoud, B. Delalat, S. Fleicher, E. Fine, T. Guliakhmedova, R. Elnathan, A. Nyska, N.H. Voelcker, M. Gozin, S. Richter, Jellyfish-based smart wound dressing devices containing in situ synthesized antibacterial nanoparticles, Adv. Funct. Mater. 29 (38) (2019) 1902783.
[142] N. Cai, C. Li, C. Han, X. Luo, L. Shen, Y. Xue, F. Yu, Tailoring mechanical and antibacterial properties of chitosan/gelatin nanofiber membranes with Fe_3O_4 nanoparticles for potential wound dressing application, Appl. Surf. Sci. 369 (2016) 492–500.
[143] C. Liu, J. Guo, X. Yan, Y. Tang, A. Mazumder, S. Wu, Y. Liang, Antimicrobial nanomaterials against biofilms: an alternative strategy, Environ. Rev. 25 (2) (2016) 225–244.
[144] Y. Feng, L. Liu, J. Zhang, H. Aslan, M. Dong, Photoactive antimicrobial nanomaterials, J. Mater. Chem. B 5 (44) (2017) 8631–8652.
[145] C. Chambers, S. Stewart, B. Su, H. Jenkinson, J. Sandy, A. Ireland, Silver doped titanium dioxide nanoparticles as antimicrobial additives to dental polymers, Dent. Mater. 33 (3) (2017) e115–e123.
[146] C.H. Woo, Y.C. Choi, J.S. Choi, H.Y. Lee, Y.W. Cho, A bilayer composite composed of TiO_2-incorporated electrospun chitosan membrane and human extracellular matrix sheet as a wound dressing, J. Biomater. Sci., Polym. Ed. 26 (13) (2015) 841–854.
[147] K. Ghosal, C. Agatemor, Z. Špitálsky, S. Thomas, E. Kny, Electrospinning tissue engineering and wound dressing scaffolds from polymer-titanium dioxide nanocomposites, Chem. Eng. J. 358 (2019) 1262–1278.
[148] R. Augustine, H.N. Malik, D.K. Singhal, A. Mukherjee, D. Malakar, N. Kalarikkal, S. Thomas, Electrospun polycaprolactone/ZnO nanocomposite membranes as biomaterials with antibacterial and cell adhesion properties, J. Polym. Res. 21 (3) (2014) 347.
[149] Y. Chen, W. Lu, Y. Guo, Y. Zhu, Y. Song, Electrospun gelatin fibers surface loaded ZnO particles as a potential biodegradable antibacterial wound dressing, Nanomaterials 9 (4) (2019) 525.

[150] W.-S. Cho, R. Duffin, M. Bradley, I.L. Megson, W. MacNee, J.K. Lee, J. Jeong, K. Donaldson, Predictive value of *in vitro* assays depends on the mechanism of toxicity of metal oxide nanoparticles, Part. Fibre Toxicol. 10 (1) (2013) 55.
[151] R. Shi, H. Geng, M. Gong, J. Ye, C. Wu, X. Hu, L. Zhang, Long-acting and broad-spectrum antimicrobial electrospun poly (ε-caprolactone)/gelatin micro/nanofibers for wound dressing, J. Colloid Interface Sci. 509 (2018) 275–284.
[152] L.A. Asri, M. Crismaru, S. Roest, Y. Chen, O. Ivashenko, P. Rudolf, J.C. Tiller, H.C. van der Mei, T.J. Loontjens, H.J. Busscher, A shape-adaptive, antibacterial-coating of immobilized quaternary-ammonium compounds tethered on hyperbranched polyurea and its mechanism of action, Adv. Funct. Mater. 24 (3) (2014) 346–355.
[153] G.Q. Blantocas, A.S. Alaboodi, A.-b.H. Mekky, Synthesis of chitosan–TiO_2 antimicrobial composites via a 2-step process of electrospinning and plasma sputtering, Arab. J. Sci. Eng. 43 (1) (2018) 389–398.
[154] A. Ehterami, M. Salehi, S. Farzamfar, A. Vaez, H. Samadian, H. Sahrapeyma, M. Mirzaii, S. Ghorbani, A. Goodarzi, *in vitro* and *in vivo* study of PCL/COLL wound dressing loaded with insulin-chitosan nanoparticles on cutaneous wound healing in rats model, Int. J. Biol. Macromol. 117 (2018) 601–609.
[155] Z. Hadisi, J. Nourmohammadi, S.M. Nassiri, The antibacterial and anti-inflammatory investigation of Lawsonia inermis-gelatin-starch nano-fibrous dressing in burn wound, Int. J. Biol. Macromol. 107 (2018) 2008–2019.
[156] I. Liakos, L. Rizzello, H. Hajiali, V. Brunetti, R. Carzino, P. Pompa, A. Athanassiou, E. Mele, Fibrous wound dressings encapsulating essential oils as natural antimicrobial agents, J. Mater. Chem. B 3 (8) (2015) 1583–1589.
[157] M. Adeli-Sardou, M.M. Yaghoobi, M. Torkzadeh-Mahani, M. Dodel, Controlled release of lawsone from polycaprolactone/gelatin electrospun nano fibers for skin tissue regeneration, Int. J. Biol. Macromol. 124 (2019) 478–491.
[158] F. Costa, I.F. Carvalho, R.C. Montelaro, P. Gomes, M.C.L. Martins, Covalent immobilization of antimicrobial peptides (AMPs) onto biomaterial surfaces, Acta Biomater. 7 (4) (2011) 1431–1440.
[159] H. Mahmoodian, O. Moradi, B. Shariatzadeh, Grafting chitosan and polyHEMA on carbon nanotubes surfaces: “Grafting to” and “Grafting from” methods, Int. J. Biol. Macromol. 63 (2014) 92–97.
[160] J.-B.D. Green, T. Fulghum, M.A. Nordhaus, Immobilized antimicrobial agents: a critical perspective, Chem. Rev. 109 (11) (2009) 5437–5527.
[161] A.R. Murphy, D.L. Kaplan, Biomedical applications of chemically-modified silk fibroin, J. Mater. Chem. 19 (36) (2009) 6443–6450.
[162] A. Peschel, How do bacteria resist human antimicrobial peptides? Trends Microbiol. 10 (4) (2002) 179–186.
[163] R.C. Goy, S.T. Morais, O.B. Assis, Evaluation of the antimicrobial activity of chitosan and its quaternized derivative on *E. coli* and *S. aureus* growth, Revista Brasileira de Farmacognosia 26 (1) (2016) 122–127.
[164] V. Saharan, A. Mehrotra, R. Khatik, P. Rawal, S.S. Sharma, A. Pal, Synthesis of chitosan based nanoparticles and their *in vitro* evaluation against phytopathogenic fungi, Int. J. Biol. Macromol. 62 (2013) 677–683.
[165] M. Arkoun, F. Daigle, M.-C. Heuzey, A. Ajji, Mechanism of action of electrospun chitosan-based nanofibers against meat spoilage and pathogenic bacteria, Molecules 22 (4) (2017) 585.
[166] S.-C. Park, C. Choi, G.-W. Jeong, H.-S. Lee, S.-J. Choi, W.-S. Kim, J.-W. Nah, Algicidal effects of free-amine water-soluble chitosan to marine harmful algal species, J. Ind. Eng. Chem. 34 (2016) 139–145.
[167] S. Chirkov, The antiviral activity of chitosan, Appl. Biochem. Microbiol. 38 (1) (2002) 1–8.
[168] Y.-J. Jeon, S.-K. Kim, Effect of antimicrobial activity by chitosan oligosaccharide N-conjugated with asparagine, J. Microbiol. Biotechnol. 11 (2) (2001) 281–286.
[169] B. Xiao, Y. Wan, M. Zhao, Y. Liu, S. Zhang, Preparation and characterization of antimicrobial chitosan-N-arginine with different degrees of substitution, Carbohydr. Polym. 83 (1) (2011) 144–150.
[170] L. Lin, A. Perets, Y. Har-el, D. Varma, M. Li, P. Lazarovici, D.L. Woerdeman, P.I. Lelkes, Alimentary ‘green’ proteins as electrospun scaffolds for skin regenerative engineering, J. Tissue Eng. Regen. Med. 7 (12) (2013) 994–1008.
[171] Y.X. Seow, C.R. Yeo, H.L. Chung, H.-G. Yuk, Plant essential oils as active antimicrobial agents, Crit. Rev. Food Sci. Nutr. 54 (5) (2014) 625–644.
[172] S. Bulman, G. Tronci, P. Goswami, C. Carr, S. Russell, Antibacterial properties of nonwoven wound dressings coated with Manuka honey or methylglyoxal, Materials 10 (8) (2017) 954.
[173] A. Oryan, E. Alemzadeh, A. Moshiri, Biological properties and therapeutic activities of honey in wound healing: a narrative review and meta-analysis, J. Tissue Viability 25 (2) (2016) 98–118.
[174] S. Martinotti, E. Ranzato, Honey, wound repair and regenerative medicine, J. Funct. Biomater. 9 (2) (2018) 34.
[175] A.M. Scagnelli, Therapeutic review: Manuka honey, J. Exot. Pet Med. 2 (25) (2016) 168–171.
[176] T. Sato, G. Miyata, The nutraceutical benefit, part iii: honey, Nutrition 16 (6) (2000) 468–469.
[177] R. Cooper, L. Jenkins, A. Henriques, R. Duggan, N. Burton, Absence of bacterial resistance to medical-grade manuka honey, Eur. J. Clin. Microbiol. Infect. Dis. 29 (10) (2010) 1237–1241.
[178] R. Yaghoobi, A. Kazerouni, Evidence for clinical use of honey in wound healing as an anti-bacterial, anti-inflammatory anti-oxidant and anti-viral agent: a review, Jundishapur J. Nat. Pharm. Prod. 8 (3) (2013) 100.
[179] Y.-H. Shan, L.-H. Peng, X. Liu, X. Chen, J. Xiong, J.-Q. Gao, Silk fibroin/gelatin electrospun nanofibrous dressing functionalized with astragaloside IV induces healing and anti-scar effects on burn wound, Int. J. Pharm. 479 (2) (2015) 291–301.
[180] I. Yousefi, M. Pakravan, H. Rahimi, A. Bahador, Z. Farshadzadeh, I. Haririan, An investigation of electrospun Henna leaves extract-loaded chitosan based nanofibrous mats for skin tissue engineering, Mater. Sci. Eng.: C 75 (2017) 433–444.
[181] M.H. Farzaei, Z. Abbasabadi, M.R. Shams-Ardekani, M. Abdollahi, R. Rahimi, A comprehensive review of plants and their active constituents with wound healing activity in traditional Iranian medicine, Wounds 26 (7) (2014) 197–206.
[182] R. Sridhar, R. Lakshminarayanan, K. Madhaiyan, V.A. Barathi, K.H.C. Lim, S. Ramakrishna, Electrosprayed nanoparticles and electrospun nanofibers based on natural materials: applications in tissue regeneration, drug delivery and pharmaceuticals, Chem. Soc. Rev. 44 (3) (2015) 790–814.
[183] S. Vakilian, M. Norouzi, M. Soufi-Zomorrod, I. Shabani, S. Hosseinzadeh, M. Soleimani, L. inermis-loaded nanofibrous scaffolds for wound dressing applications, Tissue Cell 51 (2018) 32–38.
[184] N.M. Rahmoun, Z. Boucherit-Atmani, M. Benabdallah, K. Boucherit, D. Villemin, N. Choukchou-Braham, Antimicrobial activities of the henna extract and some synthetic naphthoquinones derivatives, Am. J. Med. Biol. Res. 1 (1) (2013) 16–22.
[185] S.P. Miguel, D. Simões, A.F. Moreira, R.S. Sequeira, I.J. Correia, Production and characterization of electrospun silk fibroin based asymmetric membranes for wound dressing applications, Int. J. Biol. Macromol. 121 (2019) 524–535.
[186] G. Liu, Z. Gu, Y. Hong, L. Cheng, C. Li, Electrospun starch nanofibers: recent advances, challenges, and strategies for potential pharmaceutical applications, J. Control. Release 252 (2017) 95–107.
[187] M.C. Gunde, N.D. Amnerkar, Nutritional, medicinal and pharmacological properties of papaya (*Carica papaya* Linn.): a review, J. Innov. Pharm. Biol. Sci. 3 (1) (2016) 162–169.
[188] S. Shahid, U. Fatima, Pharmacological activities of *Carica papaya* Linn, J. Basic Appl. Sci. 14 (2018) 210–216.
[189] J. Ahlawat, V. Kumar, P. Gopinath, *Carica papaya* loaded poly (vinyl alcohol)-gelatin nanofibrous scaffold for potential application in wound dressing, Mater. Sci. Eng.: C 103 (2019) 109834.
[190] S. Agarwal, J.H. Wendorff, A. Greiner, Use of electrospinning technique for biomedical applications, Polymer 49 (26) (2008) 5603–5621.